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## ERRATA

### Volume 42

P. 359, line 1, for these read the.  
P. 370, line 20, after that insert at 15°.  
P. 371, line 5, for per cent. read per cents.  
Plate XXIV, fig. 106, delete the black smudge below the nucleus.  
P. 450, in title, for PELLETIER I read PELLETIERI.  
P. 488, line 9 from below, delete bei Sporenbildung.  
P. 490, line 3, for '96 read :06.  
P. 490, line 11 from below, for :05 read '95.

### Volume 43

P. 62, line 11, for potogravure read photogravure.  
P. 71, line 24, for UNBAN read URBAN.  
P. 76, line 17, for Rother read Rothert.  
P. 146, line 13 from below, for Dawe's read Dawes'.  
P. 146, line 8 from below, for *Blasamocitrus* read *Balsamocitrus*.  
P. 146, line 3 from below, for VAN TEIGHEM read VAN TIEGHEM.  
P. 156, line 18, for obscured read observed.  
P. 210, line 21, insert comma after stouter; delete comma after rule.  
P. 213, line 2, delete l. c.  
P. 226, line 1, insert as footnote to VON SCHRENK:

SCHRENK, H. VON, Constriction of twigs by the bag-worm and incident evidence of growth pressure. Rept. Mo. Bot Garden 17:153-181. pls. 20-26, figs. 4, charts 2. 1906.

P. 296, line 1, for HARSHBURGER read HARSHBERGER.  
P. 296, line 12, after series insert of wall charts.  
P. 299, line 11 from below, for *fructicosa* read *fruticosa*.  
P. 302, line 4, for *frutiosia* read *fruticosa*.  
P. 305, line 6, after *nonperforata* delete dash.  
P. 330, line 5 from below, for establissemnt read établissement.  
P. 333, line 1 from below, for 1-6<sup>dcm</sup> read 1-6<sup>dm</sup>.  
P. 334, line 4, for 1<sup>dcm</sup> read 1<sup>dm</sup>.  
P. 357, line 18, for secretions read secretion.

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# BOTANICAL GAZETTE

JANUARY 1907

## FERTILIZATION AND EMBRYOGENY IN CEPHALOTAXUS FORTUNEI

W. C. COKER

(WITH FIVE FIGURES AND PLATE I)

The material for this study was collected at Darlington, S. C., where the plant grew normally and produced good seeds. Collections of young ovules were also made in Bonn, Germany, but they are here represented only in *fig. 1*.

Our knowledge of the spermatogenesis of *Cephalotaxus* is confined to the work of STRASBURGER (18) and of ARNOLDI (1). Twenty-seven years ago STRASBURGER described the development of the embryo quite accurately for the stages observed, but he did not secure young proembryos. ARNOLDI has described the gametophytes and proembryo, but he has overlooked certain interesting peculiarities and is in places not sufficiently clear.

Material is not yet at hand to determine the development of the young ovule in detail, and its early history will not be considered here. However, one figure is given (*fig. 1*) from material collected January 5, 1902, at Bonn, to show the interesting midwinter condition of the ovule. This is about nine and one-half months after pollination and the pollen tube has developed into a large sac which occupies a great part of the tip of the nucellus. The body cell and two vegetative nuclei are noticed near the center. In the massive lower part of the ovule is the megasporangium, not yet divided. This winter condition will at once suggest the great difference between the genera of the Taxeae in regard to the time elapsing between the critical points of pollination, fertilization, and maturation of the seeds. In both *Torreya* (COULTER and LAND, 6) and *Cephalotaxus* the

time between pollination and maturity of the seed is about eighteen months, but the relative length of time between pollination and fertilization, and fertilization and maturity is reversed in the two genera. In *Torreya* fertilization follows in about four months after pollination, while in *Cephalotaxus* about fourteen months elapse between the two events. We find, therefore, that most of the growth of the ovule follows fertilization in *Torreya*, and precedes it in *Cephalotaxus*.

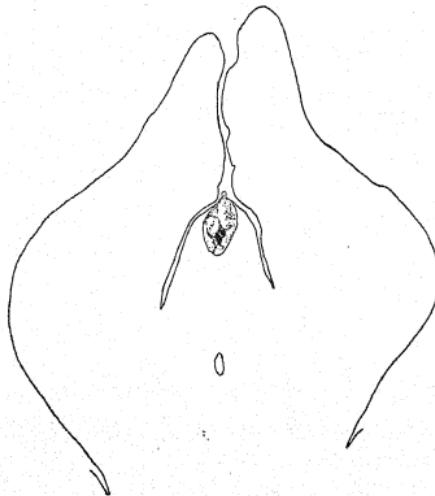


FIG. 1.—Pollinated ovule in winter condition. Bonn, Germany, Jan. 5. 1902. X45.

tion in about two months. As a result of this very rapid sequence we find that at the time of fertilization its prothallium is extremely small and delicate—more so than in any other gymnosperm, with the possible exception of *Torreya*.

The pollen grain of *Cephalotaxus* divides before being shed (STRASBURGER, 19) and there are no prothallial cells formed. The tube contents show the normal structure, the tube and stalk nuclei lying just in front of the body cell (fig. 1). ARNOLDI states that in "manchen Fällen" one can make out three nuclei in addition to the body cell in the tip of the pollen tube, and adds that, as regards the contents of the pollen tube, *Cephalotaxus* may be distinguished from

The ovule of *Cephalotaxus* at the time of fertilization is quite large, over 1.5 cm long, and the prothallium is well developed; while at the same stage, the ovule of *Torreya* is only about one-fifth as long, although at maturity it is much larger than the ovule of *Cephalotaxus*. In *Taxus* the ovule matures in one growing season, fertilization following pollination

all other conifers. These observations I cannot confirm, as all the pollen tubes examined had the usual two vegetative nuclei as in *Taxus*, *Torreya*, and other Coniferae.<sup>1</sup>

In the spring of its second season the pollen tube still grows very slowly, remaining broad and sac-like until about two or three weeks before fertilization, when it rapidly penetrates the short distance to the female prothallium and spreads its tip over the neck of an archegonium. The body cell which has been growing slowly all the time now shows the structure indicated in fig. 2.<sup>2</sup> Its protoplasm is very dense and shows well-marked radiations from a denser point just below the nucleus. The nucleus occupies a very eccentric position near the upper side of the cell, thus indicating the unequal division that is to follow.

When the tube becomes flattened out on the neck of the archegonium, the stalk and tube nuclei may be touching or slightly pressed into the body cell; usually, however, they are at a greater distance from it.

A few days before fertilization the body cell divides into two sperm cells of unequal size, the lower being in every case the larger. The difference in size is not so great as in *Torreya taxifolia* (COULTER and LAND, 6) or in *Taxus* (BELAJEFF, 3) but it is nevertheless decidedly constant. In the seventeen pollen tubes that appeared in this stage in my preparations, the difference in size of the two sperm cells could in every case be easily made out. In two of three supernumerary tubes that had been left over after fertilization, the nucleus of the smaller cell had grown larger than the other, thus obscuring to some extent the unequal distribution of the protoplasm. In figs. 3, 4, 5 are shown three pairs of sperm cells of unequal size. It will be seen that each sperm cell is quite distinct from its fellow and from the protoplasm of the pollen tube. Soon after their formation the smaller cell tends to round itself more quickly than the larger (fig. 5). Another indication that the two sperm cells are not of equivalent value is that the nucleus of the smaller may not be so dense as that of the larger (fig. 4).

<sup>1</sup> It is probable that ARNOLDI's material varied at times from the normal, as I suppose also to have been the case in my material of *Podocarpus*, where three vegetative nuclei were found in the pollen tube.

<sup>2</sup> For figs. 2-17, see PLATE I.

Each pollen tube applies itself to but one archegonium, and in fertilization only the forward and larger of the two sperm cells is functional. ARNOLDI (1) in his study of *Cephalotaxus* does not mention any difference in size between the two sperm cells. He probably overlooked it. In *Torreya taxifolia* COULTER and LAND find that the two sperm cells are very unequal and that, as in *Cephalotaxus*, each is distinct from the other. In *Torreya californica*, however, Miss ROBERTSON (17) cannot distinguish any division of the protoplasm between the two sperm nuclei. She says: "these two nuclei seem never to be surrounded by separate masses of cytoplasm, agreeing in this respect with *Pinus*." She finds that the two nuclei are of equal size and believes that only one of them is functional.

In *Taxus baccata*, according to BELAJEFF (3), STRASBURGER (19), and JAEGER (8),<sup>3</sup> the very unequal sperm cells are quite distinctly separated, and JAEGER finds their nuclei of equal size.

In my work on *Podocarpus* (4) I found that only one functional sperm cell was formed, one nucleus being thrust to the surface of the cell. In the two preparations showing this stage, this nucleus did not seem to have any separate protoplasm of its own, but I would not be sure of this without further investigation.

We find then, in summarizing our present knowledge, that all the Taxaceae so far investigated show but one functional sperm cell, and that in the Taxeae proper (with the exception of *Torreya californica*) there are two distinct sperm cells of unequal size, the difference being more pronounced in *Taxus* and *Torreya* than in *Cephalotaxus*.

In *Pinus* (Miss FERGUSON, 7), *Picea* (MIYAKE, 12), and *Abies* (MIYAKE, 13), the two sperm nuclei lie in a common protoplasm, but the forward one is the larger and alone is functional. According to MURRILL'S (14) account one would suppose that in *Tsuga* the two sperm cells are distinct, but the nucleus of the forward functional cell is decidedly larger, as in the other Abieteae above mentioned. It would seem, therefore, that in all gymnosperms whose pollen tubes fertilize but a single archegonium there is but one functional sperm cell (Taxaceae) or sperm nucleus (Abieteae).<sup>4</sup>

<sup>3</sup> JAEGER's fig. 34 shows a distinct line between the two cells, but he says: "Die Umrisse der kleinen generativen Zelle, die von Archegonium abgekährt ist, erkennen wir nur undeutlich, sie ist nicht so stark gefärbt, wie die grosse generative Zelle."

<sup>4</sup> The peculiar multiplication of the number of sperm cells found in conservatory grown material of *Cupressus Goveriana* by JUEL (Flora 93:56-62. pl. 3. 1904) is probably abnormal.

The archegonia of *Cephalotaxus* vary in number from two to five; three is a common number in my preparations. They are always situated in the micropylar end of the prothallium and are never in contact with each other for any distance and only rarely touch each other at any point. They are extremely long in comparison with their width, and are sharply pointed below. The usual shape is represented in *fig. 12*. The one shown in *fig. 9* is shorter than usual. The jacket is not nearly so well-developed as in some other conifers, and is frequently interrupted by ordinary cells (*fig. 12*).

According to ARNOLDI there are two neck cells (he mentions no exception), but in one case I found as many as five neck cells and in several cases three and four, all in one plane. In *fig. 6* a neck of three cells is shown from above. Occasionally the tip of the archegonium pushes beyond the neck cells, moving them to one side (*fig. 7*). Up to ten or fifteen days before fertilization the archegonia have very little protoplasmic contents. The nucleus at this time is small and is very close to the upper end. The protoplasm begins to thicken rapidly just before the division of the central cell, which occurs about ten days before fertilization. The ventral canal nucleus is without any distinctive protoplasm of its own, as ARNOLDI has already pointed out, resembling in this respect that of *Podocarpus* (COKER, 4), *Taxodium* (COKER, 5), *Cryptomeria* (LAWSON, 11), *Thuja* (LAND, 9), and *Juniperus* (NORÉN, 15). Miss ROBERTSON found the spindle of the division in *Torreya californica*, but no later stages. In *fig. 8* is shown the ventral nucleus and the egg nucleus soon after the division. The former is at the upper surface of the protoplasm. In *fig. 9* the canal nucleus is shown in its usual position, but there is the curious abnormality of two other nuclei in the egg. The canal nucleus sometimes moves away from the surface and approaches nearer the egg nucleus (*fig. 10*), but this must be considered an abnormality. The canal nucleus generally disappears before fertilization and cannot be demonstrated at that time.

In Darlington, S. C., fertilization took place, in 1903, from the fifth to the eighth of May. The necks of the archegonia are at this time at the bottom of the pits formed by upgrowths of the prothallium. The pollen tubes reach the archegonia before these pits are formed and the advancing prothallium grows around them. In case

there is no pollen tube above an archegonium, the growth of the prothallium may close the pit and bury the archegonium completely.

Both sperm cells are discharged into the egg, but the two vegetative nuclei may remain behind. The larger sperm cell advances to the egg nucleus, the sperm nucleus sinks into it, and the sperm protoplasm gradually surrounds the fusing nuclei, exactly as I have already described for *Taxodium*. *Figs. 11, 12, 13, 14* show stages of approach and fusion. The second sperm cell remains above (*figs. 11, 12*) and may approach pretty close to the fusion nucleus (*fig. 14*). ARNOLDI (1) says that soon after the sperm cells enter the archegonium, the protoplasm becomes mixed with that of the egg. He does not observe the sperm protoplasm investing the fusion nucleus.

The contribution of the sperm protoplasm to the proembryo has also been observed in *Torreya* by Miss ROBERTSON (17) and COULTER and LAND (6), in *Cryptomeria* by ARNOLDI (1) and LAWSON (11), in *Juniperus* by NORÉN (15), and in *Sequoia* by ARNOLDI (2). In LAWSON's (10) work on *Sequoia* he does not confirm ARNOLDI, but finds that only a very small amount of the sperm protoplasm enters the egg with the nucleus, the rest remaining behind in the pollen tube. This odd behavior is similar to the process of fertilization in *Taxus* as described by BELAJEFF (3). There is no starch in the sperm cells of *Cephalotaxus* such as is found in *Taxodium*, *Sequoia*, *Cryptomeria*, and *Juniperus*.

The first division of the fusion nucleus occurs near the center of the archegonium. The spindle is very small compared with the size of the nucleus and is entirely intranuclear. *Fig. 14* shows an early stage in this division. The sperm protoplasm has not yet entirely invested the nucleus. In *fig. 15* the division is complete and the two nuclei are approaching the base of the archegonium. Two extra nuclei are shown above—one is probably the second sperm nucleus. The next division occurs before the proembryo has reached the base of the archegonium. Three of the four nuclei produced by this division are shown in *fig. 16*.

In *fig. 17* is shown the eight cell-stage which now occupies the base of the archegonium. Five of the eight nuclei are in the section. These eight nuclei now divide again simultaneously, and after the formation of the sixteen daughter nuclei, cell walls are formed for

the first time. *Fig. 18* shows this stage just at the beginning of the cell wall formation. The cells are not arranged in regular tiers throughout, being most irregular in the central region. At the upper end are two fairly even tiers, and the tip is occupied nearly always by two superimposed cells. In archegonia with more rounded bases, which appear occasionally, there may be two cells side by side in the tip.

After the formation of cell walls the sixteen cells divide again to form thirty-two, and it seems probable that the upper tier divides horizontally to form the rosette cells and suspensors. I have not found any case, however, where the rosette consists of free nuclei, as is so common in conifers. The cells of the more or less regular tier above the suspensors are enclosed in the cell walls, and occasionally one or more of them may divide horizontally. In *fig. 19* the thirty-two cell-stage is represented. The upper tier (*r*), which answers to the rosette of five cells; the next tier (*s*), which forms the suspensors, also contains five cells; while the remaining cells are not in regular tiers except near the tip, where there are three distinct tiers, the upper of two cells, the two lower of one cell each.

The suspensors now begin to elongate and cell divisions occur in any of the cells of the middle region below the suspensors and above the two tip cells. These divisions are no longer simultaneous, but occur here and there as in ordinary growth. In *fig. 20* is shown an embryo in which the suspensors have just begun to elongate. One cell of the upper tier has divided into two, and the number of cells in the middle region has increased somewhat. The two tip cells show



FIG. 19.—Thirty-two-celled proembryo just before the elongation of suspensors.  $\times 185$ .



FIG. 18.—Sixteen celled proembryo with cell walls beginning to form.  $\times 185$ .

no signs as yet of the disorganization that, according to STRASBURGER (18), they are to undergo. The protoplasm of the tip cell is somewhat less dense than the others, but not markedly so. The



FIG. 20.—Slightly older proembryo; suspensors elongating. May 19, 1903.  $\times 150$ .

A much older embryo is represented in fig. 21. Above the crumpled suspensors and embryonal tubes three abortive embryos are seen. These are probably the products of three additional archegonia, as it certainly is not the normal thing for an archegonium to produce more than one embryo in *Cephalotaxus*. In none of the proembryos seen did the suspensors show signs of separating, as they do in many other conifers, and from the structure of the proembryo this does not surprise us.

ARNOLDI (1) does not state definitely the number of cell divisions before the formation of walls in the proembryo, and from his description one would suppose the cells to be arranged in more definite tiers than they actually are. As to the rosette, my observations agree with STRASBURGER'S (18), that the cells composing it are enclosed

nuclei of this cell and the one above it, however, are much larger than the others, and the cells give every indication of being actively secretive. At this stage they have been driven some distance downward into the prothallial tissue, but instead of being crushed by the growth behind they seem to be opening the way for the progress of the embryo. As I have not yet secured stages immediately following this, I cannot determine the ultimate fate of the tip cells. It is certain, however, that they persist for some time in healthy and active condition at the tip of the embryo.

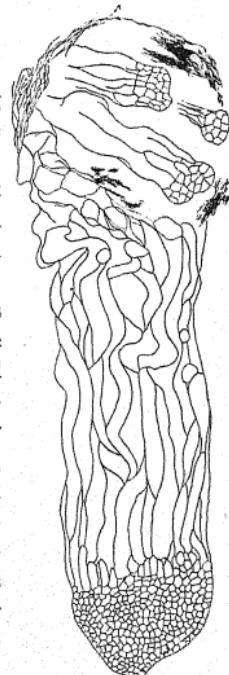


FIG. 21.—Older embryo with abortive ones above. June 16, 1903.  $\times 45$ .

in walls and may divide after their formation. In one figure he shows each rosette cell divided into a group. According to him the embryo proper is formed from the group of cells lying between the tip cell (or cells) and the suspensors, and the tip cell is destroyed.

In comparing the proembryo of *Cephalotaxus* with that of its relatives, we find that it shows some resemblance to *Taxus*, but very little to *Torreya*. In *Taxus*, according to JAEGER (8), cell walls are not formed before the sixteen cell-stage at the earliest, but, as one would expect from the shape of the archegonium, there is no long and narrow cell at the apex of the embryo. In *Torreya*, according to Miss ROBERTSON (17) and COULTER and LAND (6), cell walls are formed in the four cell-stage. In *Torreya californica* the organized proembryo consists of an exposed rosette, a tier of four or six suspensors, and a "cluster" of tip cells (Miss ROBERTSON, 17). In *T. taxifolia*, however, the proembryo entirely fills the archegonium with twelve or eighteen cells, all closed, and in this stage passes the winter (COULTER and LAND, 6). *Cephalotaxus*, *Taxus*, and *Podocarpus* are the only conifers so far examined in which cell walls are not formed in the proembryo before the sixteen cell-stage.

CHAPEL HILL, N. C.

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#### EXPLANATION OF PLATE I

FIGS. 1, 18-21 are in the text.

FIGS. 2-17 reduced one-third. Magnifications accordingly are now only two-thirds the number given.

FIG. 2. Body cell, stalk nucleus, and pollen tube nucleus in tip of a pollen tube that has reached the prothallium. April 21, 1903.  $\times 670$ .

FIGS. 3-5. Sperm cells ready for fertilization. May 5 and 8, 1903.  $\times 670$ .

FIG. 6. Neck cells seen from above.  $\times 670$ .

FIG. 7. Neck cells pushed to one side.  $\times 250$ .

FIG. 8. Ventral canal (*v*) and egg (*e*) nuclei in tip of archegonium.  $\times 370$ .

FIG. 9. Archegonium showing ventral canal nucleus and two other nuclei below.  $\times 150$ .

FIG. 10. Upper part of archegonium with egg (*e*) and ventral canal (*v*) nuclei.  $\times 370$ .

FIG. 11. Upper part of archegonium with functional sperm cell approaching egg nucleus, and second sperm nucleus above. May 5, 1903.  $\times 670$ .

FIG. 12. Archegonium and pollen tube just after fertilization; fusing nuclei in center and second sperm nucleus above.  $\times 150$ .

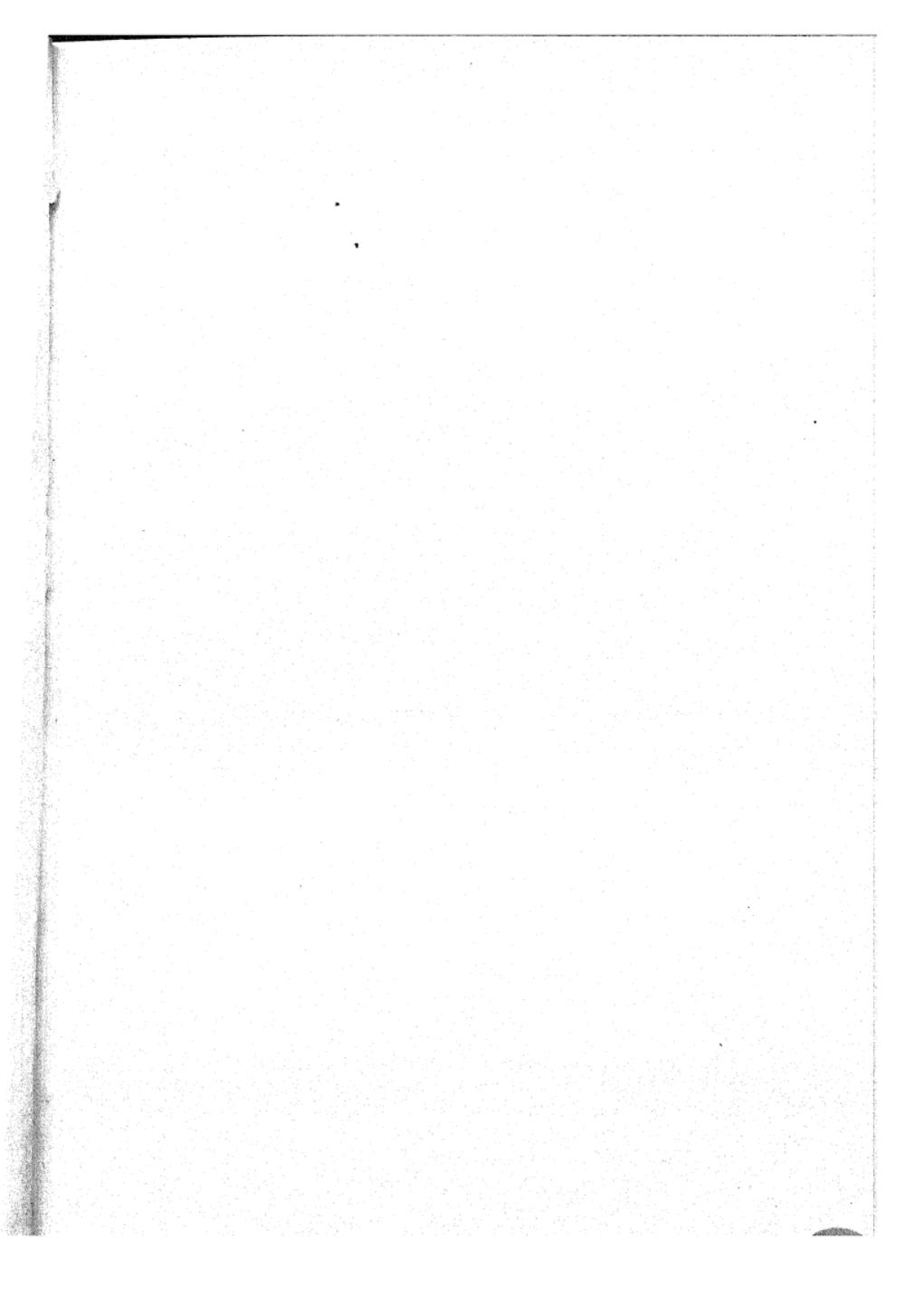
FIG. 13. Fusing nuclei with sperm protoplasm on upper side.  $\times 670$ .

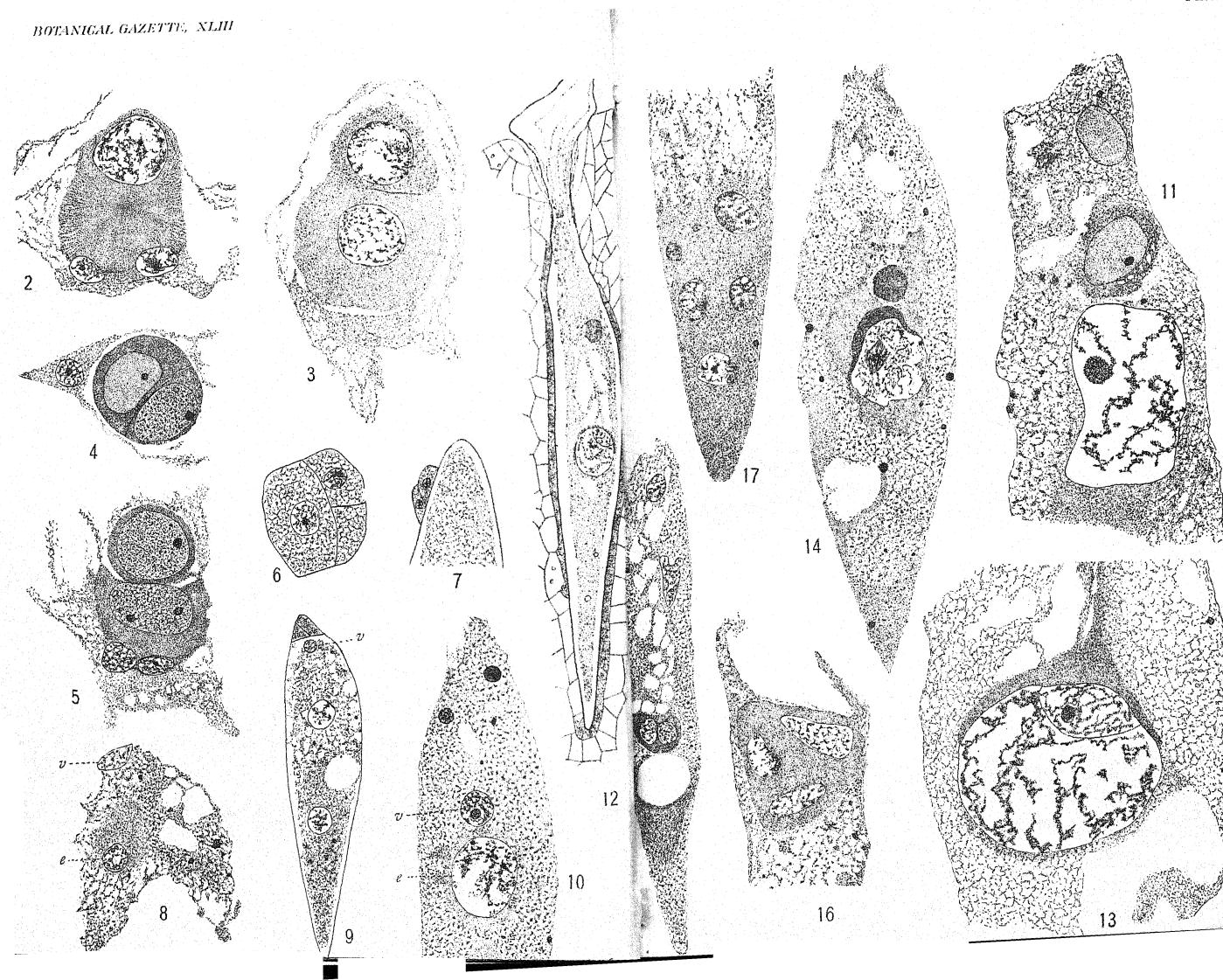
FIG. 14. Spindle of first division of fusion nucleus; second sperm nucleus above.  $\times 370$ .

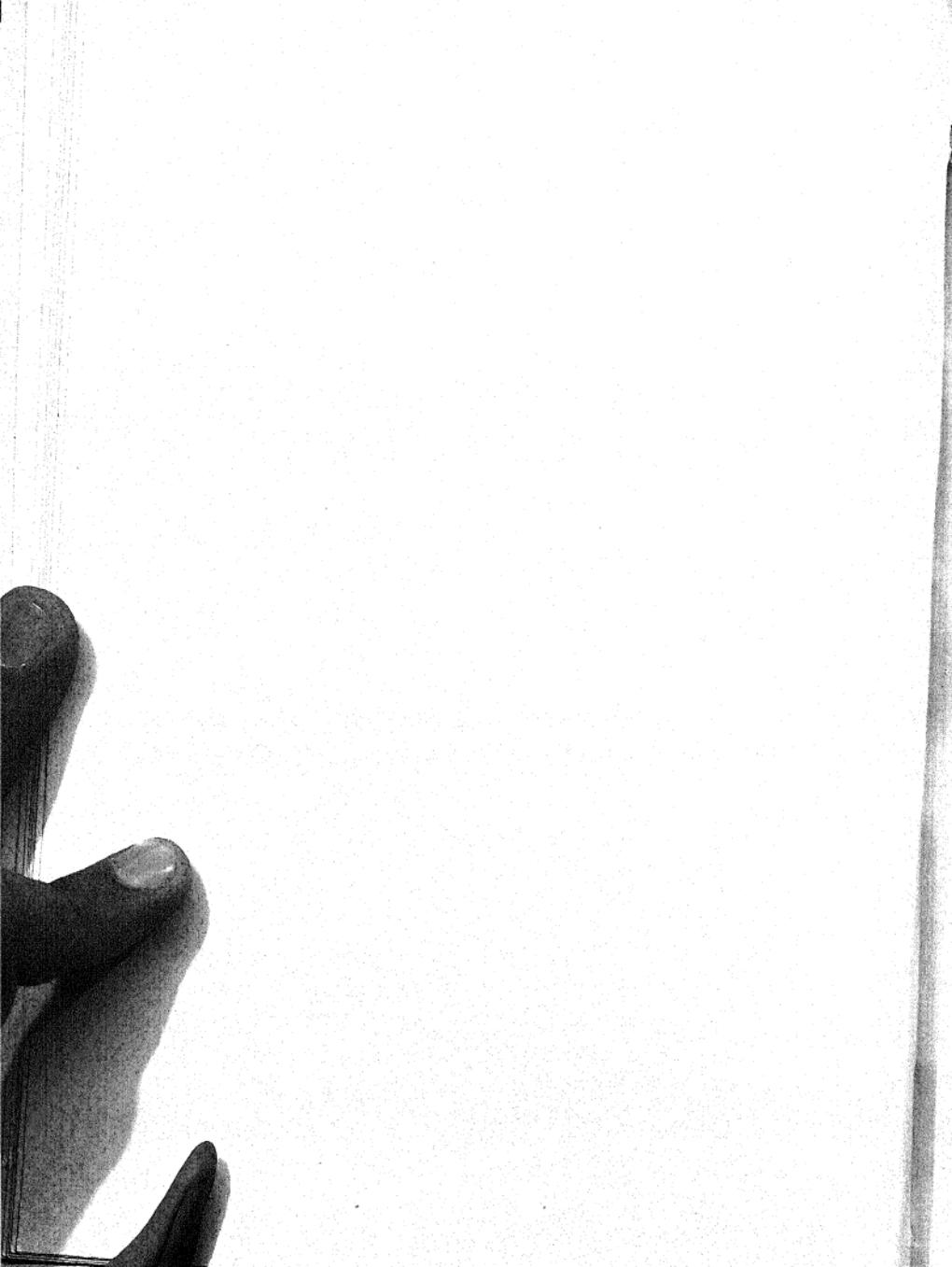
FIG. 15. Two-celled proembryo with two nuclei above. May 8, 1903.  $\times 250$ .

FIG. 16. Four-celled proembryo near base of archegonium.  $\times 670$ .

FIG. 17. Eight-celled proembryo in base of archegonium.  $\times 670$ .







TOXIC LIMITS AND STIMULATION EFFECTS OF SOME  
SALTS AND POISONS ON WHEAT  
CONTRIBUTIONS FROM THE HULL BOTANICAL LABORATORY  
LXXXVIII

G. H. JENSEN

(WITH THIRTY-FOUR FIGURES)

That the vital activities in plants may be inhibited or accelerated by many mineral salts has long been known in a general way. Hitherto the problem has been attacked mainly from the standpoint of toxic solutions. The work which has taken into consideration the influence of a poison in soil has invariably been done with native soils. In these there are always present a great variety of chemical agents which may individually or collectively react with the introduced poison, and hence no fundamental conclusion can be deduced therefrom. TRUE and OGLEVEE in 1904 (42) showed that the toxicity of a solution was reduced by the introduction of solid insoluble particles. This first suggested the possibility of comparing results obtained from plants grown in solution and in soil. The limited extent to which these authors carried their experiments, however, gave us no more than a suggestion of the possibilities. Previous experimenters, furthermore, have dealt mainly with small parts of the plants, such as the root-tips, determining their limit of endurance.

The present work was undertaken with the purpose of ascertaining the toxic limit for the whole plant in both solution and soil cultures, and thus to make comparisons of the two conditions; to ascertain the effect of pure quartz sand in raising the toxic limit for a given poison upon the same plant; to add to the knowledge of stimulation effects of poisons in solutions; to discover whether these effects were also manifested in soil cultures; and to secure evidence of comparative absorption of different poisons by the same sand.

**Historical**

The literature dealing with toxic effects on plants divides itself naturally into two classes; first, that which deals with the lower

plants; and second, that which deals with the higher plants. The studies of toxic agents on algae and fungi, while they bear upon the present paper only indirectly, frequently furnish valuable data for comparison. Unfortunately the determinations have been made with different ends in view, and have been tabulated in such a way that only here and there can the most general comparisons be made.

LIVINGSTON (23, 24, 25) has made perhaps the most valuable contribution to this work on the lower plants. His experimentation takes into consideration many toxic salts; determines their killing concentrations as well as the strengths which cause changes in vegetative character of the alga *Stigeoclonium*. He also attempts (25) to correlate the work of previous authors with each other and with his own.

Fungus growth and spore germination as affected by toxic salts in solution was studied by CLARK (8). Since it is generally recognized that  $\text{Cu}(\text{NO}_3)_2$  and  $\text{CuSO}_4$  have very nearly the same if not identical toxic limits for a given plant (25, p. 18), it is interesting to note from this table:

Aspergillus in $\text{Cu}(\text{NO}_3)_2$ , dies in $25n/100$
Sterigmatocystis in $\text{CuSO}_4$ , dies in $125n/1000$
Penicillium in $\text{Cu}(\text{NO}_3)_2$ , dies in $n/1$
Oedocephalum in $\text{Cu}(\text{NO}_3)_2$ , dies in $156n/10000$
Botrytis in $\text{Cu}(\text{NO}_3)_2$ , dies in $313n/10000$

Thus Penicillium will endure approximately four times as much copper as Aspergillus; eight times as much as Sterigmatocystis; thirty times as much as Botrytis; and sixty times as much as Oedocephalum.

According to STEVENS (39) many metallic salts affect spore production in fungi. Among these  $\text{Cu}(\text{NO}_3)_2$  and  $\text{CuSO}_4$  inhibit spore formation at a lower concentration than any of the others tried by him, the response occurring at  $n/100$  in Penicillium.

The response of acceleration of growth of plants as caused by toxic elements in solutions has been studied in conjunction with the response of death. There seems to be a general agreement among authors that zinc is the most active stimulating agent to vegetative growth. Yet the concentrations at which  $\text{ZnSO}_4$  are said to produce most marked acceleration are far from agreeing. Thus, RICHARDS

(34) says that  $6n/$  to  $12n/100000$  and ONO (32) that  $25n/$  to  $200n/100000$  produce the greatest dry weight of mycelium in *Aspergillus*. RICHTER (35) did not get acceleration of growth with  $\text{CuSO}_4$  nor with  $\text{AgCl}$ .

Within the past decade considerable work on seedlings of higher plants dealing with their relation to toxic agents has appeared. In nearly all cases this work has been limited to the effect of the toxic agent upon some particular part or organ, and has taken no account of the endurance of the plant as a whole for the poison. Thus KAHLENBERG and TRUE (21) record a large number of experiments upon white lupine radicles. They determined for this organ the fatal dose of various organic and inorganic substances. They found that H and OH ions are the deleterious agents. This work establishes the limit of endurance of the root-tip of white lupine only, and not, as COUPIN (10) points out, the endurance of the whole plant. In the same fashion HEALD (19) worked with seedlings of *Zea*, *Pisum*, and *Cucurbita*. He found that in  $\text{KCN}$ ,  $\text{K}_4\text{Fe}(\text{CN})_6$ , and  $\text{K}_3\text{Fe}(\text{CN})_6$  the electro-negative ion or anion was toxic. In all others it was the electro-positive ion or cation. PLOWMAN (33) dealt with the question from the standpoint of electricity and magnetism. From the fact that plants in the soil in the vicinity of the positive pole were stimulated, while those at the negative pole were not, he concluded that ionization of the soil solutions bore directly upon growth.

That roots may accommodate themselves to gradually increasing concentrations of toxic substances (38) has long been known, as has also the fact that the power of absorbing water containing salt is lost more and more by roots (36) the longer they are in contact with it. GUTHRIE and HELMS (16) worked with wheat, maize, barley, and rye in pot cultures, using native soil. Their idea was to determine the endurance of some farm crops for certain injurious substances frequently present in soils. Table A, made from their data, shows the per cent. of poison which prevents germination.

From these results it would seem that seeds will successfully germinate in somewhat higher concentration than that in which the plants will later grow. TRUE and OGLEVEE (42) found that various insoluble substances, potato starch, paraffin, coal, glass, filter paper,

TABLE A

Reagent	Wheat	Maize	Barley	Rye
NaCl	Germination prevented by .	o.20%	o.50%	o.25%
	Growth prevented by . . . .	o.20	o.25	o.20
Na <sub>2</sub> CO <sub>3</sub>	Germination prevented by .	o.5-1.0	o.50	o.60
	Growth prevented by . . . .	o.40	o.25	o.40
NH <sub>4</sub> CNS	Germination prevented by .	o.015	o.02+	....
	Growth prevented by . . . .	o.005	o.02+	....
NaClO <sub>3</sub>	Germination prevented by .	o.053	o.006+	o.007
	Growth prevented by . . . .	o.003	o.004	o.006
AsO(OH) <sub>3</sub>	Germination prevented by .	o.5+	o.80+	o.60*
	Growth prevented by . . . .	o.10	o.60	o.10

\* Not effective.

when added in finely divided particles to toxic solutions, markedly reduced the toxicity. They account for the reduction as being due to adsorption of poisonous particles by the substances introduced.

In a recent paper LIVINGSTON (27) goes very fully into the physiological properties of a remarkably sterile soil, known locally as Tacoma Park soil. He concludes that some reagents, such as pyrogallol, calcium carbonate, and tannic acid, may act chemically upon the toxic bodies of the soil, while ferric hydrate and carbon black are merely absorbents of the same.

KANDA (22) worked with solution cultures, and pot cultures of native soil. The latter were watered with poisonous solutions of the same salts from which his solution cultures were made. He was the first to show that ZnSO<sub>4</sub> is an active stimulant to higher plants. He did not observe stimulation with solution cultures of CuSO<sub>4</sub>, but soil cultures gave quite marked stimulation. The results that he records for *Pisum* and *Vicia Faba* in this respect agree with my own on wheat.

HARTER (18) exposed wheat roots for twenty four hours to toxic solutions. He found that varieties of wheat from arid regions of Russia, where the soil is alkaline, are more resistant than are those grown in more humid regions. The reagents used by HARTER were sodium carbonate, bicarbonate, sulfate, and chlorid; and magnesium sulfate and chlorid. His work therefore naturally suggests that relative toxicity of salts must be learned by experimenting upon a single variety or strain; and to overcome errors due to individual variation many plants must be used.

That it is next to impossible to coordinate data from different authors is evident from Table B, taken from CAMERON and BREAZEALE (7). For, as they say, the toxicity needed "to kill a seedling or disorganize an entire radicle varies widely from that required to completely hinder any growth, or that which will permit some elongation."

TABLE B

SOLUTE	TOXIC LIMITS FOR	
	Whole seedling	Tip of radicle
H <sub>2</sub> SO <sub>4</sub> .....	n/9	n/15000
HCl.....	n/10	n/18000
HNO <sub>3</sub> .....	n/7	n/15000
Acetic acid.....	n/13	n/20000
Oxalic acid.....	n/1	n/15000
Succinic acid.....	n/1	n/20000
Calcium chlorid.....	n/3	n/4
Calcium nitrate.....	n/3	n/5

DANDENO (11) worked with pea, lupine, and corn. He found that the toxicity of poisonous solutions is reduced by the presence of pure quartz sand. Thus in CuSO<sub>4</sub>, seedlings will grow in n/4096 when sand is present, while they will stand only n/32768 when no sand is present. He also found that the effect is related to the size of the particles, more growth being permitted with fine sand than with coarse sand. My own experimentation with H<sub>2</sub>SO<sub>4</sub> shows similar results. Thus with number 5/o, the finest quartz, growth was inhibited at 7n/100 to 6n/100; in number 2/o, the next coarser grade, at 5n/100 to 4n/100; in number 2, a rather coarse sand, at 4n/100 to 2n/100; in number 4, a very coarse grade, at 2n/100 to 9n/1000; in solution (H<sub>2</sub>O), at 8n/1000 to 6n/1000. Which goes to show that the coarser the sand the less the same amount of solution is reduced in toxicity. Or, in other words, the coarser the sand, the nearer does the culture approach to solution media. DANDENO does not think with TRUE that the growth of lateral roots in solutions that killed the primary radicle is due to "accommodation," but that the first growth of the radicle has reduced the toxicity to such an extent that the later roots can endure it. That the quantity of toxic agent has considerable to do with the effect upon growth, he shows

by a series of eight cultures, successively planted in different quantities of poison. Thus, seedlings lived in  $1^{\text{cc}}$  HCl  $n/1024$  for the first day, and not in quantities more than  $1^{\text{cc}}$ ; while at the end of the eight successive plantings in the same solutions seedlings were able to survive on the eighth day in  $12^{\text{cc}}$  HCl  $n/1024$ ; hence definitely proving that each successive planting reduces the toxicity of the solution. He says this is due to exudations of substances from the root. These same exudations furnish organic material for the growth of fungi, which (as was also my experience) are very apt to make their appearance in a few days.

BREAZEALE (5) made soil extracts of unproductive soils. To these extracts, filtered through a Chamberland filter he added various solids, such as ferric hydrate, calcium nitrate, calcium carbonate, carbon black, and fine quartz flour. He found that ferric hydrate and carbon black produce abnormal lengthening of roots, while the quartz flour does not. In his experiments on maize with sulfuric acid  $n/2750$  to  $n/3250$  prevented growth. Quartz flour did not change the apparent death limit. Clear sand, bits of filter paper, and paraffin shavings showed similar results with  $\text{H}_2\text{SO}_4$ . His results thus far, therefore, are diametrically opposed to those of TRUE and OGELVEE. With  $\text{CuSO}_4$ , on the other hand, carbon black reduced the toxic effect. My own results, in so far as quartz flour is concerned, showed almost invariably that it reduces toxicity. The only exception is in phenol and alcohol.

#### Materials

Ground quartz, or quartz flour, of the grade commercially known as 5/o, which is a very fine powder, was the soil medium used. Wire baskets  $8 \times 8 \text{ cm}^2$ , paraffined and covered as described by LIVINGSTON (27), formed the pots for the soil cultures.

For the solution series,  $500^{\text{cc}}$  bottles with a wide mouth were generally used. A smaller dark bottle, of  $70^{\text{cc}}$  capacity, was also used, but did not give as uniformly good results as the larger bottle.

The wheat used throughout these experiments was obtained from the Bureau of Soils, Washington, D. C., through the kindness of Dr. LIVINGSTON. It is the variety known as *Chul*, grown in Utah, and was brought from central Turkestan by Dr. ERNST BESSEY.

All salts used for nutrient media, as well as those used as poisons, were Kahlbaum's C. P. grade.

The ordinary distilled water of the laboratory was re-distilled in glass flasks. From this redistilled water all solutions were made.

#### Methods

Preliminary experiments for the determination of the optimum water content of the quartz were first carried out with redistilled water and with nutrient solution. It was found that 12 to 15 per cent. by weight of water was best.

The wheat seeds were germinated in sphagnum and in quartz. It was found that the sphagnum was more satisfactory, because foreign particles could be more easily removed from the roots at transplanting. Transplanting was done when plants had attained a maximum height of 3<sup>cm</sup>.

Three hundred and thirty-six grams of quartz were weighed out for each culture basket. This was mixed with 42<sup>cc</sup> of solution, thus making 12.5 per cent. of moisture content. This damp soil was then packed in the basket, and with the culture series completed, all were planted with plants as nearly alike as possible. The plants (five in each pot) stood in a straight line and protruded through a slit in the paraffined paper cover. The latter was then sealed to the sides of the pot with melted paraffin. The pot was labeled with the strength of solution it contained and the weight. The slit through which the plants protruded was loosely plugged with ordinary cotton, so that the loss of weight was practically reduced to the transpiration by the plants. After two or three days the pots were put on the scale-pan, the original registered weight was placed on the opposite pan, and a balance obtained by adding to each some solution of its own strength from a pipette. Thus the weight was again made what it was at the time of planting. This was done as often as needed; at first each two or three days; afterward, as plants got larger, every day. In this way the amount of water was never more than 12.5 per cent. and probably never less than 10 per cent. The amounts added were recorded, and their sum constitutes the total transpiration for a pot as recorded in the tables. At the end of the experiment, usually in 15 to 30 days, the longest

sprout of each plant was measured, the recorded length being taken from the surface of the pot to the tip of the leaf. The average of these lengths for a given pot constitutes the average length of sprout recorded for that culture. The plants were then cut off close to the surface, cut into short pieces into a crucible, and the green weight recorded. After drying in a Bausch and Lomb oven for 5 to 7 days at 100-110° C. the dry weight was recorded.

Corks fitting the bottles used for solution cultures were perforated with a small cork-borer so that six plants could be grown in each of the larger bottles. The corks were then boiled for several hours, with numerous changes of water, in order to remove any possible injurious ingredients. After being dried, they were boiled in paraffin, to insure an upper surface that could not be wetted. The wheat plants were now inserted through the perforations, so that the seed and roots were below the lower surface of the cork and in the solution. The holes were loosely plugged with ordinary cotton to fasten the plants and to prevent evaporation. Bottles were weighed from time to time and solution replaced from a pipette, record being kept of the amount lost by transpiration from each bottle. The length of the sprout was measured from the upper surface of the cork to the tip of the longest leaf of each plant as in the case of the soil cultures. The green and dry weight of the whole plant from the solution cultures was always taken; while in the soil cultures only the aerial portion is available for comparison.

Fungus contamination in solution cultures was practically overcome by sterilizing seeds in a 2 per cent. CuSO<sub>4</sub> solution for three-fourths of an hour; washing afterwards in sterilized water; planting in sterilized sphagnum, and transplanting in a sterile chamber into sterilized solutions. The sterile chamber was a box 100×50×50<sup>cm</sup>, with a glass cover and a glass window in front. Rubber-cloth sleeves which fit snugly around the wrists of the operator were provided. The chamber was thoroughly sprayed with CuSO<sub>4</sub> before using.

The series in solution and in soil were grown at the same time, under the same conditions of light, heat, and other external factors.

#### Experimentation

Responses to toxic salts are of two kinds: (a) acceleration and (b) retardation of growth which progresses as the concentration in-

creases until the death point is reached. By the *death point* is not meant that concentration at which all elongation of root or sprout from the very first is inhibited, but that which kills the plant when the stored food of the seed has been exhausted. Thus it was found that after 5 to 10 days plants which at first grew well lost their vigor and turned yellow.

The criteria used for comparing the effect of the substances tested are: first, the total transpiration from each pot or culture; second, the average length of the sprout; third, the green weight; fourth, the dry weight of plants in a pot.

The curves are as follows: The unbroken line is the average length of the sprout; the broken line is the total transpiration; the dotted line is the green weight; and the dot-and-dash line represents the curve of the dry weight.

#### COPPER SULFATE

In the solution series plants all die in  $n/1000$  strength and do very badly in  $5n/100000$ . There is no indication of any stimulation, as the control in distilled water has grown more than any other set. In the series where  $50^{\text{gm}}$  of 5/o quartz sand is added to an equal weight of solution, that is, where the quartz stood half way up the column of liquid, we find the wheat growing in  $n/1000$  and not altogether dead in  $4n/1000$ . The toxicity has evidently been reduced. In the series where  $90^{\text{gm}}$  quartz to  $25^{\text{gm}}$  solution is used, i. e., the solution stands about a centimeter above the surface of the sand, the toxic effect has been still further reduced. The killing point now lies between  $4n/1000$  and  $8n/1000$ . In the soil series, i. e., where 12.5 per cent. of solution is present, the death point lies between  $n/100$  and  $3n/100$ . It will be evident, therefore, that the greater the ratio of quartz to the poisonous solution, provided this does not exceed the optimum water content for growth, the less the toxic effect of the copper sulfate. *Fig. 1* represents a  $\text{CuSO}_4$  series in soil six days after planting. The series is close, running from left to right  $n/10$ ,  $9n/100$ ,  $8n/100$ ,  $7n/100$ ,  $6n/100$ ,  $5n/100$ ,  $4n/100$ ,  $3n/100$ ,  $2n/100$ ,  $n/100$ ,  $9n/1000$ ,  $8n/1000$ ,  $7n/1000$ ,  $6n/1000$ ,  $5n/1000$ ,  $4n/1000$ ,  $3n/1000$ ,  $2n/1000$ ,  $n/1000$ ,  $n/2500$ ,  $n/5000$ ,  $n/7500$ ,  $n/10000$ , control. It shows that up to  $3n/100$  all elongation is inhibited. Beyond this point it shows a rather uniform increase

of growth, as the solution strength, with which the soil is moistened, decreases. Normal growth is reached at about  $2n/1000$ . Between the point at which no growth takes place and where normal growth

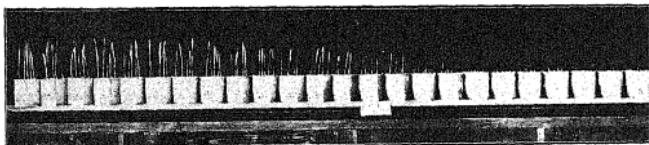


FIG. 1.—Copper sulfate series in soil. Aug. 24–30. 1905.

may be attained, we will, for convenience, call the tension line or line of stress. It means that region of concentration in which a struggle for life is apparent. In *fig. 2*, which is a photograph of the same series 12 days old arranged in opposite direction, it will be noted

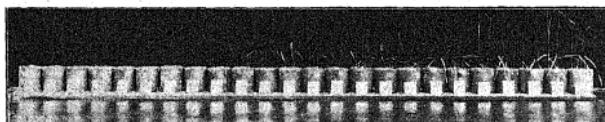


FIG. 2.—Copper sulfate series in soil. Aug. 24–Sept. 26. 1905.

that  $n/1000$  to  $n/2500$  has grown more than the control. Also that  $n/100$  and  $9n/1000$ , which were growing in *fig. 1*, are now dead. It thus shows that during the first six days the seedling was able to endure a higher concentration than afterward; and further, that

acceleration does not occur until after the reserve food of the seed has been exhausted.

The solution series represented in *fig. 3* had concentrations from left to right as follows:

$9n/1000$ ,  $8n/1000$ ,

$7n/1000$ ,  $6n/1000$ ,  $5n/1000$ ,  $4n/1000$ ,  $3n/1000$ ,  $2n/1000$ ,  $n/1000$ ,  $n/2500$ ,  $n/5000$ ,  $n/7500$ ,  $n/10000$ , control. In this series no

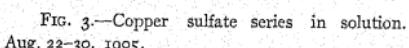


FIG. 3.—Copper sulfate series in solution. Aug. 22–30. 1905.

elongation has taken place in concentrations above  $3n/1000$ ; the curve of increase of growth rapidly rises as the concentration decreases, and much more rapidly than it does in soil. There is no indication of any acceleration in the photographed series, nor was any acceleration found in  $\text{CuSO}_4$  solution culture, although the experiment was twice repeated with dilutions as low as  $n/100000$ .

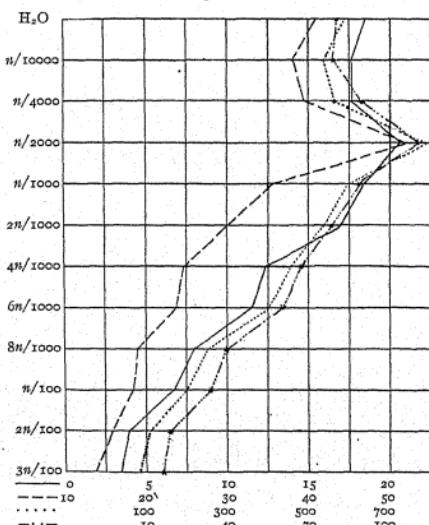


FIG. 4.—Copper sulfate series in soil. Feb. 17–Mar. 3, 1906.

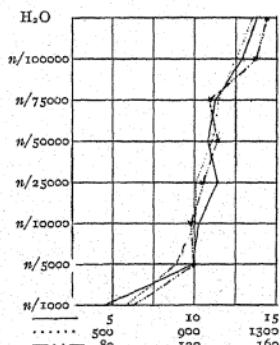


FIG. 5.—Copper sulfate series in solution. Feb. 20–Mar. 3, 1906.

As has already been stated, the fatal concentration of  $\text{CuSO}_4$  in solution cultures is approximately one tenth that of the fatal concentration in soil; or, the quartz has reduced the toxic effect about nine tenths.

Tables I, II, III follow and curves (figs. 4, 5) precede. From these it will be seen that the transpiration, the length of sprout, and the green and dry weights vary quite uniformly.

TABLE I  
 $\text{CuSO}_4$  SERIES IN SOIL; 12.5% MOISTURE. FEBRUARY 17–MARCH 3, 1906

	H <sub>2</sub> O	$n/10000$	$n/4000$	$n/2000$	$n/1000$	$2n/1000$	$4n/1000$	$6n/1000$	$8n/1000$	$n/100$	$2n/100$	$3n/100$
Av. length of sprout, cm....	18.96	17.76	17.70	20.68	18.44	16.96	11.98	11.56	8.02	7.26	4.10	3.42
Total transpiration, gm....	41.6	37.9	39.4	52.0	35.4	30.2	24.8	23.8	18.5	18.5	15.9	13.9
Total green weight, mg....	590	535	563	782	597	530	450	400	250	200	115	92
Total dry weight, mg....	80	78	88	112	90	79	68	62	40	35	18	17

TABLE II  
 $\text{CuSO}_4$  SERIES. EXPERIMENTS TO SHOW THE EFFECT OF QUARTZ IN SOLUTION

	H <sub>2</sub> O	$\frac{n}{100000}$	$\frac{n}{1000}$	$\frac{4n}{1000}$	$\frac{8n}{1000}$	$\frac{n}{100}$	$\frac{2n}{100}$
50gm quartz to 50cc solution.....							
Avg. length of sprout, cm.....	15.35	11.42	6.57	3.60	3.12		
50gm quartz to 25cc solution.....							
Avg. length of sprout, cm.....	14.85	14.72	11.65	7.2	4.8	3.4	2.2

TABLE III  
 $\text{CuSO}_4$  SERIES IN SOLUTION; 500cc BOTTLES. FEBRUARY 20-MARCH 3, 1906

	H <sub>2</sub> O	$\frac{n}{100000}$	$\frac{n}{75000}$	$\frac{n}{50000}$	$\frac{n}{25000}$	$\frac{n}{10000}$	$\frac{n}{5000}$	$\frac{n}{1000}$
Average length of sprout, cm.....	13.87	12.95	11.33	10.98	11.41	10.25	9.58	4.06
Total green weight, mg.....	1206	1123	980	982	915	931	825	575
Total dry weight, mg.....	155	150	128	130	123	118	120	90

#### LEAD NITRATE

In the preliminary experiment with lead nitrate from which fig. 6 is taken, the series runs from right to left as follows:  $8n/100$ ,

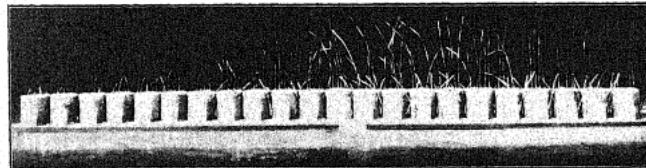


FIG. 6.—Lead nitrate series in soil. Sept. 2-14. 1905.

$7n/100$ ,  $6n/100$ ,  $5n/100$ ,  $4n/100$ ,  $3n/100$ ,  $2n/100$ ,  $n/100$ ,  $9n/1000$ ,  $8n/1000$ ,  $7n/1000$ ,  $6n/1000$ ,  $5n/1000$ ,  $4n/1000$ ,  $3n/1000$ ,  $2n/1000$ ,  $n/1000$ ,  $n/2500$ ,  $n/5000$ ,  $n/7500$ ,  $n/10000$ , control.

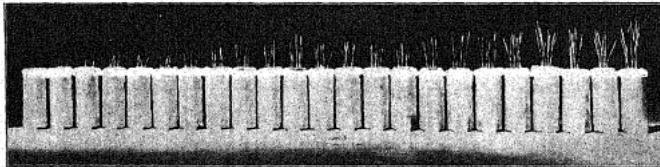


FIG. 7.—Lead nitrate series in solution. Sept. 10-14. 1905.

The same concentrations, arranged in the same order, hold for the series in solution, fig. 7.

The experiment with soil was thrice repeated, and the results of each experiment are indicated in the curves, *figs. 8, 9, 10*. The separate tables are not given, but Table IV embodies the averages obtained from the three, as does also the curve, *fig. 11*. The greatest stimulation, at  $6n/1000$ , falls off in concentrations both above and below this. If the control be considered "normal growth," it will

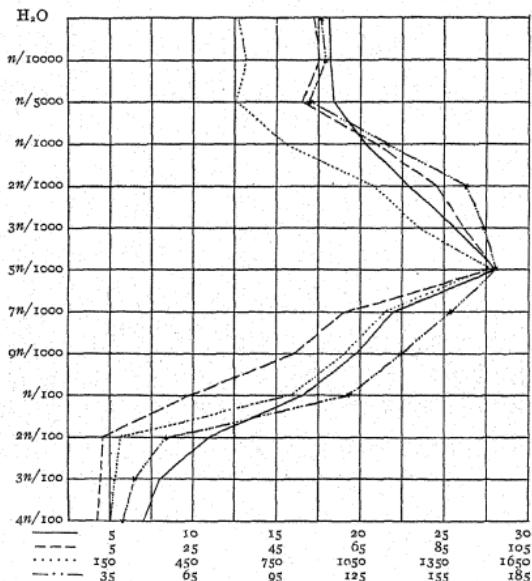
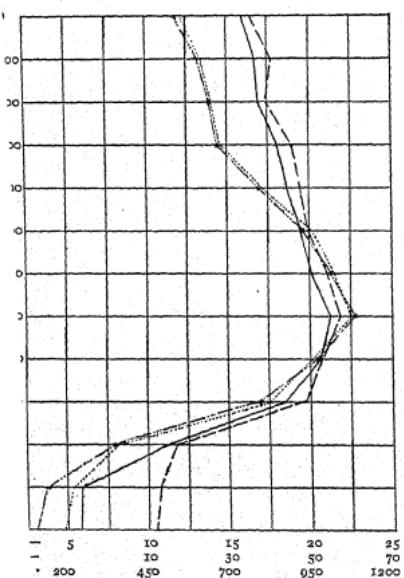


FIG. 8.—Lead nitrate series in soil. Nov. 19-Dec. 20. 1905.

be evident by reading *fig. 11* that all concentrations from  $n/100$  to  $n/10000$  inclusive cause acceleration. Death occurs at  $4n/100$ , a very slight elongation is shown at  $3n/100$ , and a little more at  $2n/100$ . Between  $2n/100$  and  $n/100$  a rapid increase in growth rate occurs. In comparison with  $\text{CuSO}_4$ , therefore, acceleration may be caused by concentrations very near the fatal concentration.

In the solution series of lead nitrate, Table V and *fig. 12*, the death point lies at  $4n/100$  to  $2n/100$ , approximately at the same concentration as in soil. Hence there is little if any indication of reduction of toxic effect, in so far as the death point is concerned.

Acceleration is not evident in nearly as marked a degree as in soil, however, and is not evident in nearly as concentrated solutions. Hence, while no appreciable change in the killing strength is made evident by the presence of quartz, a very marked change has occurred with respect to acceleration. In solution acceleration occurs at  $n/5000$  to  $n/10000$ .



-Lead nitrate series in soil. Feb. 2-19. 1906.

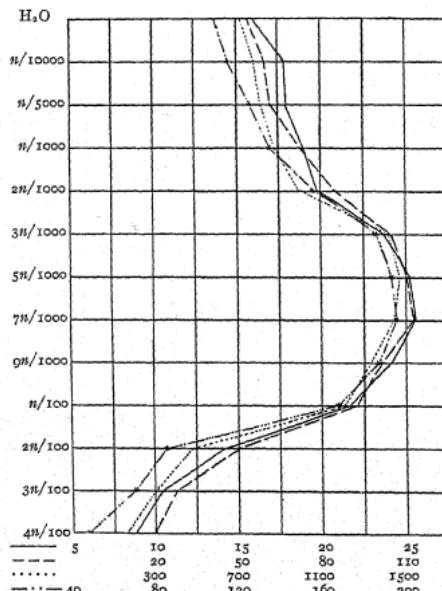


FIG. 10.—Lead nitrate series in soil. Apr. 10-27. 1906.

TABLE IV  
Pb(NO<sub>3</sub>)<sub>2</sub> SERIES IN SOIL. AVERAGE OF THREE EXPERIMENTS

	H <sub>2</sub> O	$\frac{n}{10000}$	$\frac{n}{5000}$	$\frac{n}{1000}$	$\frac{2n}{1000}$	$\frac{3n}{1000}$	$\frac{5n}{1000}$	$\frac{7n}{1000}$	$\frac{9n}{1000}$	$\frac{n}{100}$	$\frac{2n}{100}$	$\frac{3n}{100}$	$\frac{4n}{100}$
Av. length of sprout, cm.	16.61	17.87	17.01	19.46	20.46	21.15	21.54	23.18	21.61	10.34	12.21	8.31	5.77
Total transpiration, gm.	48.00	52.77	55.83	62.57	72.30	71.40	81.80	87.60	65.06	55.60	17.47	14.33	11.30
Total green weight, mg...	608	682	688	776	950	1100	1125	1173	1069	984	336	295	180
Total dry weight, mg...	97	105	110	117	134	135	150	140	140	130	63	47	43

TABLE V  
 $\text{Pb}(\text{NO}_3)_2$  SERIES IN SOLUTION. FEBRUARY 2-17. 1906

	$\text{H}_2\text{O}$	$n/10000$	$n/5000$	$n/1000$	$2n/1000$	$4n/1000$	$6n/1000$	$8n/1000$	$n/100$	$2n/100$	$4n/100$
Av. length of sprout, cm.	10.5	12.47	11.35	10.2	10.4	0.75	8.15	8.0	8.02	7.3	5.3
Total transpiration, gm.	11.6	12.30	12.50	11.6	11.1	9.00	9.20	9.7	9.7	7.0	7.0
Total green weight, mg...	670	725	640	620	585	540	523	520	476	445	408
Total dry weight, mg....	85	95	84	85	79	77	78	75	73	72	70

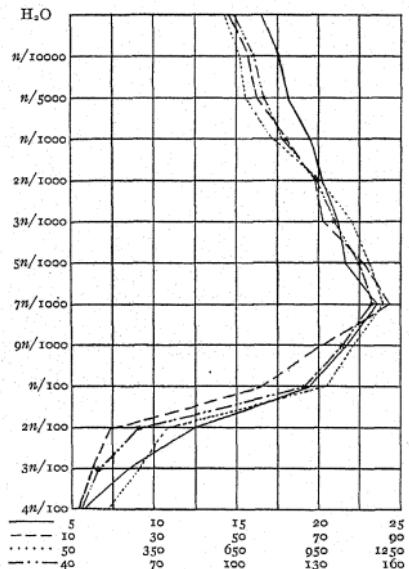


FIG. 11.—Lead nitrate. Average of three experiments.

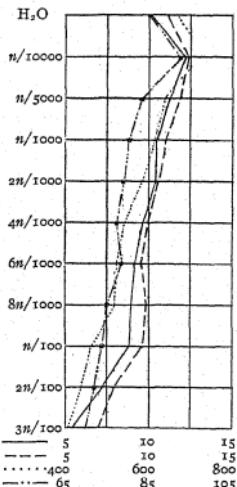


FIG. 12.—Lead nitrate series in solution. Feb. 2-17. 1906.

#### SILVER NITRATE

The experiments with silver nitrate were repeated three times. *Figs. 14, 15* are taken from the first one. The results of the different tests correspond quite closely, hence only the averages are here given in Table VI and graphically in *fig. 13*.

The toxic limit is about the same concentration ( $4n/100$  to  $3n/100$ ) as with lead nitrate; there is very little toxic effect shown above  $9n/1000$ ; the region of acceleration is at  $2n/1000$ .

TABLE VI  
 $\text{AgNO}_3$  SERIES IN SOIL. AVERAGE OF THREE EXPERIMENTS

	$\text{H}_2\text{O}$	$n$ 10000	$n$ 5000	$2n$ 1000	$3n$ 1000	$5n$ 1000	$6n$ 1000	$8n$ 1000	$9n$ 1000	$n$ 100	$2n$ 100	$3n$ 100	$4n$ 100
Av. length of sprout, cm.	20.64	21.50	22.48	22.58	21.44	20.06	10.68	10.74	17.82	16.42	15.50	13.22	2.55
Total transpiration, gm..	61.4	62.2	66.9	67.9	65.6	57.6	54.9	53.2	51.1	42.1	36.1	30.5	16.1
Total green weight, mg..	845	840	897	934	864	822	800	790	770	713	350	220	35
Total dry weight, mg..	130	136	139	150	147	144	137	131	130	122	110	97	15

In fig. 14 is represented a series of 23 cultures six days old, in quartz, running from left to right  $n/10$ ,  $9n/100$ ,  $8n/100$ , etc., as in the case of  $\text{CuSO}_4$  and  $\text{Pb}(\text{NO}_3)_2$ . A small amount of elongation has taken place in  $4n/100$ , considerable in  $3n/100$ , and in  $2n/100$  the growth is about normal. In fig. 15 the same series is 12 days old.

Here  $4n/100$  and  $3n/100$  are dead, while  $2n/100$  has maintained its rate of growth almost equal with the control. The death limit is therefore very clearly pointed out. On the other hand, acceleration is very vague.

The solution series (fig. 16) represents a series beginning at the left with  $7n/1000$ ,  $6n/1000$ ,  $5n/1000$ , etc., as in figs. 14, 15. Here

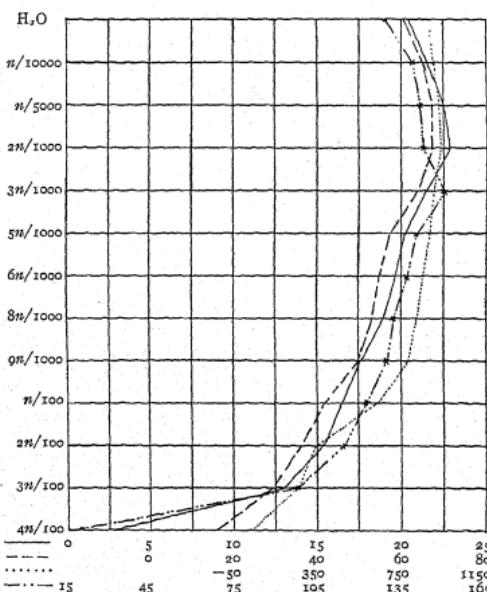


FIG. 13.—Silver nitrate series in soil. Average of results.

all elongation is inhibited in concentrations up to  $4n/100$ , and from  $4n/100$  to  $n/1000$  there is a slight elongation, with considerable

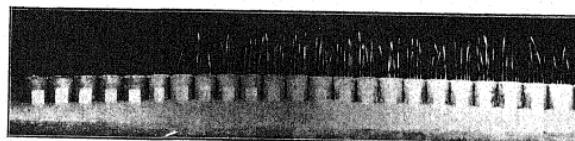


FIG. 14.—Silver nitrate series in soil. Sept. 10-15. 1905.

broadening of the leaves. In concentrations from  $n/1000$  to  $n/10000$  elongation increases. That actual acceleration might be definitely

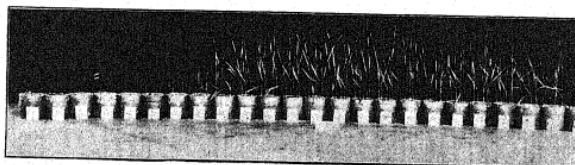


FIG. 15.—Silver nitrate series in solution. Sept. 10-22. 1905.

shown, however, another series with solutions more dilute was grown. In this the series runs as follows:  $n/10000$ ,  $n/25000$ ,  $n/50000$ ,

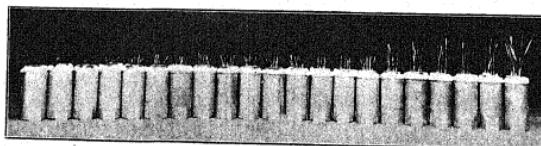


FIG. 16.—Silver nitrate series in solution. Sept. 10-15. 1905.

$n/100000$ , control. A definite region of acceleration is shown in fig. 17, which represents the results obtained with these very dilute solutions. See also Table VII.

TABLE VII  
AgNO<sub>3</sub> SERIES IN SOLUTION. FEBRUARY 19-MARCH 4. 1906

	H <sub>2</sub> O	$\frac{n}{100000}$	$\frac{n}{75000}$	$\frac{n}{50000}$	$\frac{n}{25000}$	$\frac{n}{10000}$
Av. length of sprout, cm.....	0.25	0.51	10.76	11.16	7.33	5.73
Total green weight, mg.....	870	899	965	981	887	572
Total dry weight, mg.....	109	112	117	121	90	84

It is evident that the quartz has a very marked effect in reducing the toxic effect of the silver. For example, the death point in soil is at a concentration of  $3n/100$  to  $2n/100$ ; in solution the death point is at  $n/10000$ ; this, in comparison with other poisons used, is a considerable difference.

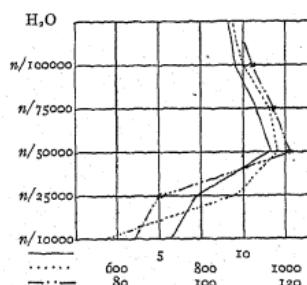


FIG. 17.—Silver nitrate series in solution. Feb. 19-Mar. 4. 1906.

varying quantity of quartz, was set up. The set consisted of seven  $500\text{cc}$  bottles. The solution strength used was  $5n/1000$ . The amounts of solution and quartz sand in each bottle were as follows:

Number	1	2	3	4	5	6	7
Solution	$150\text{cc}$	$208\text{cc}$	$310\text{cc}$	$365\text{cc}$	$410\text{cc}$	$455\text{cc}$	$500\text{cc}$
5/o quartz	625gm	625gm	500gm	375gm	250gm	125gm	25gm

That is, we have in 1 about 19 per cent. solution to 81 per cent. sand; in 2, 25 per cent. to 75 per cent. sand; in 3, 38 per cent. to 62 per cent. sand or supersaturated; in 4 the solution stands about  $2\text{cm}$  above the sand; in 7 the sand stands less than  $1\text{cm}$  from the bottom; 5 and 6 are intermediate between 4 and 7. The experiment lasted ten days and the results obtained are as follows:

TABLE VIII  
TO SHOW EFFECT OF VARYING AMOUNTS OF QUARTZ IN POISONED SOLUTION

No.	Length of sprout.						Average	Remarks
1	1.5	1.2	1.6	0.8	1.4	0.7	1.2	Roots all dead
2	2.5	2.3	2.2	1.6	1.3	1.0	1.8	Roots all dead
3	3.1	3.2	3.2	1.5	2.6	0.7	2.4	Roots dead and very slightly elongated
4	2.5	2.2	2.6	2.6	2.5	3.3	2.6	Roots had penetrated into liquid but were dead
5	3.0	3.7	3.0	2.5	2.9	2.1	2.86	Roots alive, plants dead
6	8.8	11.6	10.7	9.2	10.4	11.7	10.4	Plants healthy
7	11.7	10.4	11.2	10.1	9.0	10.5	10.5	Plants healthy

The data show an increasing growth with the increase in amount of sand present. This agrees with TRUE and OGLEVEE's results. At the end of the experiment the solutions in which the plants had grown were carefully titrated with NaCl, giving the following results:

1. Sand about 1<sup>cm</sup> high, considerable precipitate.
2. Sand about 3.5<sup>cm</sup> high, some precipitate.
3. Sand about 6<sup>cm</sup> high, minute trace.
4. Sand about 8<sup>cm</sup> high, no trace.
5. Sand about 10<sup>cm</sup> high, no trace.
6. Sand filled bottle, no trace.
7. Sand filled bottle, no trace.

I endeavored to make quantitative determinations, and thus definitely to settle the absorptive power of a given quantity of quartz, and at the same time the relation of the amount of silver to length of plant grown therein. My attempts, however, were unsuccessful.

The solutions from the above experiment were filtered free from sand, and again used for solution culture in small black bottles (70°c) with the following results:

1. Gave no elongation of root or stem. Roots and base of stem dark brown.
2. Stems slightly elongated. Leaves very much etiolated. Roots long and healthy.
3. About like 2.
4. Stems healthy. Leaves green. Roots white, long and healthy. Av. size 7.9<sup>cm</sup>.
5. Same as in 4. Average length of sprout 8.3<sup>cm</sup>.
6. Soil extract from previous experiment insufficient for this test.
7. Soil extract from previous experiment insufficient for this test.

In order to test still further the absorptive power of the quartz, a series of cultures, each containing the same quantity of sand and solution, were set up. The difference here was that each culture had a different strength of solution. The quantity of sand used for each was 270 grams, and of solution 400 grams. The concentrations are indicated in table IX.

TABLE IX  
EXPERIMENT TO SHOW EFFECT OF QUARTZ UPON DIFFERENT CONCENTRATIONS  
OF  $\text{AgNO}_3$

	$\frac{5\text{H}}{100}$	$\frac{n}{100}$	$\frac{n}{500}$	$\frac{n}{2500}$	$\frac{n}{12500}$	$\frac{n}{62500}$
Average length of sprout, cm...	3.1*	5.3	8.5	9.4	10.2	13.0
Total green weight, mg.....	255	230	388	500	545	565
Total dry weight, mg.....	67	47	60	80	82	117

\*Dead.

In comparison with this, a set of the same concentrations of solution, in the same kind of bottle ( $500^{\circ}\text{C}$ ) but containing no sand, were set up. The plants were grown the same length of time, in the same external conditions, and gave the following results: In  $n/62500$  and  $n/12500$  good growth was obtained, but in all solutions of greater concentration plants all died. It is quite evident, therefore, that the quartz added reduced the toxic effect of  $n/100$ ,  $n/500$ , and  $n/2500$  to something less than  $n/2500$ ; perhaps, as is shown by the first series of solution cultures, to something less than  $n/10000$ . This conclusion is also drawn from the fact that roots, stems, and leaves were as vigorous in  $n/2500 + \text{quartz}$  as were the plants in  $n/12500$  solution alone.

After filtering away sand the solutions were again used in solution cultures in small black bottles ( $70^{\circ}\text{C}$ ) with the following results:

	$\frac{n}{100}$	$\frac{n}{100}$	$\frac{n}{500}$	$\frac{n}{2500}$	$\frac{n}{12500}$	$\frac{n}{62500}$
Average length of sprout, cm...	0.0	4.5	10.9	12.2	11.3	11.3

This shows that it is not necessarily the presence of the quartz *at the time of growth* which reduces the toxicity, but that quartz having stood in the solution has taken out some of the poison. Here, as in a previous experiment, the titration failed to yield quantitative results. The conclusion, however, is inevitable that quartz does remove considerable quantities of silver nitrate from solution, or at least renders it inert.

#### ZINC SULFATE

The series shown in *figs. 18, 19* has the same arrangement and

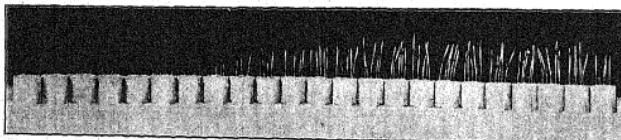


FIG. 18.—Zinc sulfate series in soil. Sept. 19-25. 1905.

concentration of  $\text{ZnSO}_4$  as that shown and explained in *figs. 15, 16* for  $\text{AgNO}_3$ .

Unfortunately, the plate showing the condition of the plants at the end of the experiment—18 days growth—was spoiled. However, by referring to Table X and *fig. 20* which represents this later condition, it will be seen that there is acceleration in concentrations  $5n/10000$  to  $n/10000$ . In comparison with lead nitrate this acceleration is in weak solutions and is not nearly as well marked as in the  $Pb(NO_3)_2$ . The series in solution of  $ZnSO_4$  (*figs. 19, 21*, and

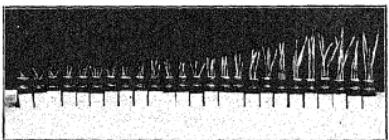


FIG. 19.—Zinc sulfate series in solution. Sept. 19-25, 1905.

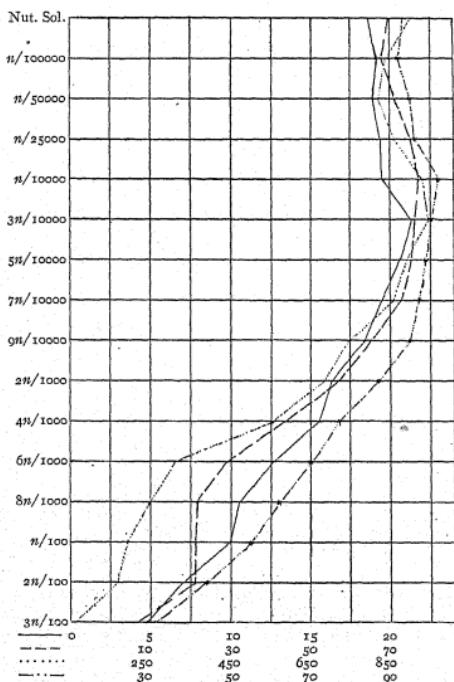


FIG. 20.—Zinc sulfate series in soil. Oct. 21-Nov. 18. 1905.

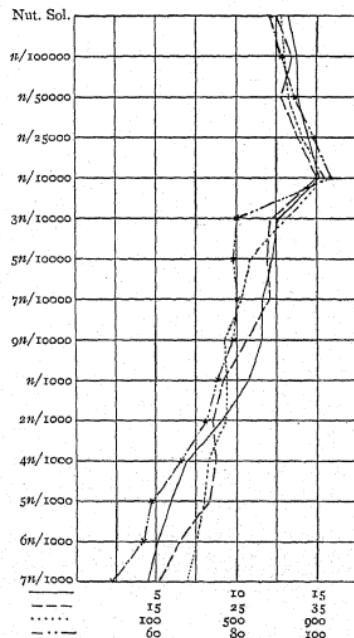


FIG. 21.—Zinc sulfate series in solution. Oct. 14-Nov. 14. 1905.

Table XI) does not show acceleration. It was thought that this was due to too strong solutions, hence the experiment was twice repeated with the concentrations as dilute as  $n/100000$ , as is seen in Table XI and fig. 21, but no very decided acceleration was obtained. This is contrary to the results obtained by RICHARDS (34), who found that  $ZnSO_4$  was a most powerful stimulant to growth in fungi. ONO (32) and RICHTER (35) also report very strong stimulation with zinc sulfate. These observations correspond more nearly to my results obtained with soil.

It is very interesting to note the relatively small reduction of toxicity by the quartz in  $ZnSO_4$  as compared with silver nitrate.

TABLE X  
 $ZnSO_4$  SERIES IN SOIL. OCTOBER 21-NOVEMBER 18, 1905

	Nut. sol.	$\frac{n}{10000}$	$\frac{n}{50000}$	$\frac{n}{25000}$	$\frac{n}{10000}$	$\frac{3n}{10000}$	$\frac{5n}{10000}$	$\frac{7n}{10000}$
Average length of sprout, cm.....	18.76	19.90	18.86	19.56	19.60	21.42	20.60	19.50
Total transpiration, gm.....	70.00	67.90	71.20	75.50	76.80	76.80	75.20	72.50
Total green weight, mg.....	905	847	810	857	929	949	890	890
Total dry weight, mg.....	94	92	95	96	102	100	99	97

	$\frac{9n}{10000}$	$\frac{2n}{1000}$	$\frac{4n}{1000}$	$\frac{6n}{1000}$	$\frac{8n}{1000}$	$\frac{n}{100}$	$\frac{2n}{100}$	$\frac{3n}{100}$
Average length of sprout, cm.....	18.28	16.20	15.42	12.72	10.30	10.12	7.14	4.9
Total transpiration, gm.....	64.60	56.00	43.80	29.20	21.90	21.40	21.	6.7
Total green weight, mg.....	748	677	601	310	250	190	169	76
Total dry weight, mg.....	96	87	77	70	62	55	44	31

TABLE XI  
 $ZnSO_4$  SERIES IN SOLUTION. OCTOBER 14-NOVEMBER 14, 1905

	Nut. sol.	$\frac{n}{100000}$	$\frac{n}{50000}$	$\frac{n}{25000}$	$\frac{n}{10000}$	$\frac{3n}{10000}$	$\frac{5n}{10000}$	$\frac{7n}{10000}$
Average length of sprout, cm.....	13.20	13.85	13.83	14.70	15.25	12.77	12.20	11.45
Total transpiration, gm.....	30.80	32.70	30.50	32.30	35.90	29.40	29.20	29.70
Total green weight, mg.....	717	740	746	782	948	730	567	527
Total dry weight, mg.....	89	91	94	100	104	80	79	80

	$\frac{9n}{10000}$	$\frac{n}{1000}$	$\frac{2n}{1000}$	$\frac{4n}{1000}$	$\frac{5n}{1000}$	$\frac{6n}{1000}$	$\frac{7n}{1000}$
Average length of sprout, cm.....	11.47	10.57	9.10	6.9	5.97	5.02	4.57
Total transpiration, gm.....	26.30	23.80	21.80	22.40	21.80	18.60	15.40
Total green weight, mg.....	445	443	447	369	354	315	307
Total dry weight, mg.....	76	77	77	66	59	57	47

## IRON NITRATE

It was found impracticable to grow plants in the nutrient solution with which the experimentation was begun on account of precipitation of one or more of the salts by such reagents as  $\text{AgNO}_3$ ,  $\text{Pb}(\text{NO}_3)_2$ ,

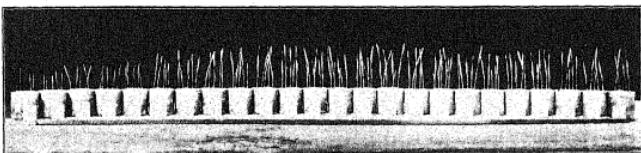


FIG. 22.—Iron nitrate series in soil. Sept. 15-21. 1905.

etc. Hence it was decided to grow them in distilled water, with no other element present than the poison. In order to test whether results obtained by both methods were comparable, two series, one

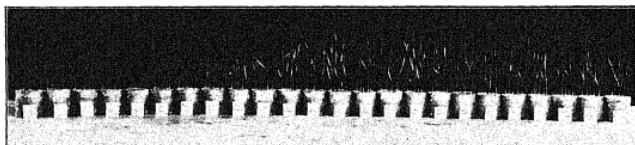


FIG. 23.—Iron nitrate series in soil. Sept. 15-27. 1905.

in water, the other in nutrient media, to each of which the poison had been added in the same proportions, were set up side by side. The experimentation with  $\text{Fe}_2(\text{NO}_3)_6$  proved that water grows the plants as well as does the nutrient solution during the time of the experiment. The curve of maximum growth is as great and comes at the same concentration with both. The killing concentrations also coincide.

The greatest growth in solution cultures is at  $n/2500$  to  $n/7500$ , while in soil cultures it occurs at  $7n/1000$  to  $3n/1000$ . Hence there

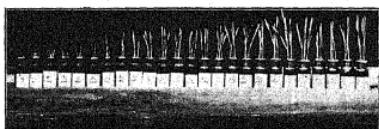


FIG. 24.—Iron nitrate series in solution. Sept. 15-21. 1905.

is a considerable reduction of stimulating effect caused by the presence of the quartz. Again the killing strength in solution is about  $n/100$ , while in soil it is from  $6n/100$  to  $4n/100$ .

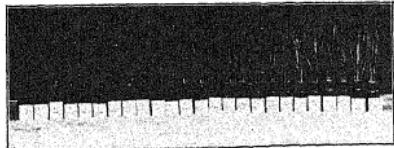


FIG. 25.—Iron nitrate series in solution. Sept. 15-27, 1905.

poisonous elements in the latter as in the former. *Fig. 22* shows the  $\text{Fe}_2(\text{NO}_3)_6$  soil series after growing five days. It will be noticed that some elongation has taken place in the culture farthest to the left, which is  $n/10$ . Further,

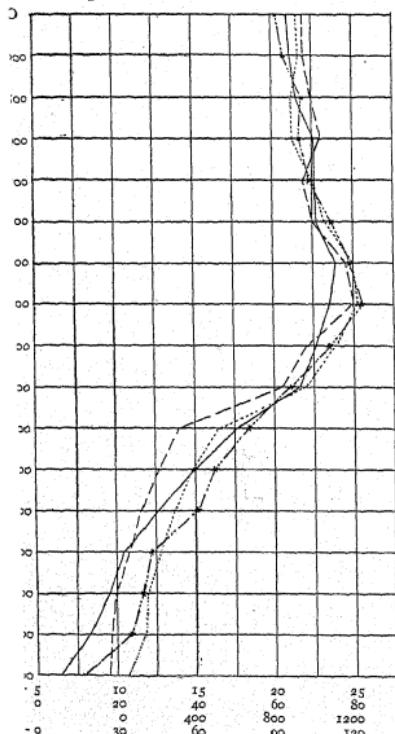


FIG. 26.—Iron nitrate series in soil with nutritive n. Oct. 14-Nov. 8, 1905.

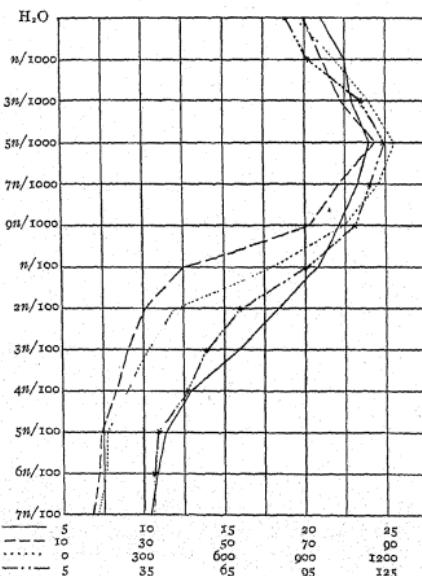


FIG. 27.—Iron nitrate series in soil without nutritive solution. Oct. 29-Nov. 24, 1905.

it is also clear, that no very marked stimulation above control, at extreme right, is perceptible. In *fig. 23*, which is the same series photographed when twelve days old, plants in pots from  $n/10$  to  $5n/100$  are dead, and the region of active stimulation is quite evident, lying at  $9n/1000$  to  $3n/1000$ . In *figs. 24, 25* the same thing is illustrated for the water cultures with  $\text{Fe}_2(\text{NO}_3)_6$  at the same ages as in *figs. 22, 23*. Tables XII and XIII give comparative results with and without nutrient media in the soil. The same is graphically shown in curves *figs. 26, 27*.

TABLE XII  
 $\text{Fe}_2(\text{NO}_3)_6$  SERIES IN SOIL WITH NUTRIENT SOLUTION. OCTOBER 14—  
 NOVEMBER 8, 1905

	Nut. sol.	$\frac{n}{1000}$	$\frac{n}{7500}$	$\frac{n}{5000}$	$\frac{n}{2500}$	$\frac{n}{1000}$	$\frac{3n}{1000}$	$\frac{5n}{1000}$	$\frac{6n}{1000}$	$\frac{7n}{1000}$
Av. length of sprout, cm...	21.10	21.16	21.52	22.62	22.60	23.5	23.9	23.5	22.7	
Total transpiration, gm...	67.6	68.8	70.7	72.8	68.6	70.5	78.7	80.0	68.6	
Total green weight, mg...	945	902	905	914	1010	1010	1202	1217	1098	
Total dry weight, mg....	91	95	98	99	101	112	118	123	110	
	$\frac{9n}{1000}$	$\frac{n}{100}$	$\frac{2n}{100}$	$\frac{3n}{100}$	$\frac{4n}{100}$	$\frac{5n}{100}$	$\frac{6n}{100}$	$\frac{7n}{100}$		
Av. length of sprout, cm.....	21.6	17.8	12.7	12.7	10.4	9.5	8.3	6.4		
Total transpiration, gm.....	65.7	36.0	28.4	26.1	23.1	19.0	18.3	18.1		
Total green weight, mg.....	987	520	301	310	223	145	135	64		
Total dry weight, mg.....	99	80	67	65	43	41	36	17		

TABLE XIII  
 $\text{Fe}_2(\text{NO}_3)_6$  SERIES IN SOIL WITHOUT NUTRIENT SOLUTION. OCTOBER 29—NOVEMBER 24, 1905

	H <sub>2</sub> O	$\frac{n}{1000}$	$\frac{3n}{1000}$	$\frac{5n}{1000}$	$\frac{7n}{1000}$	$\frac{9n}{1000}$	$\frac{n}{100}$	$\frac{2n}{100}$	$\frac{3n}{100}$	$\frac{4n}{100}$	$\frac{5n}{100}$	$\frac{6n}{100}$	$\frac{7n}{100}$
Av. length of sprout, cm...	21.08	22.60	22.84	23.92	23.12	22.04	20.96	18.44	15.90	12.90	11.36	10.94	10.38
Total transpiration, gm...	70.0	72.2	77.0	88.5	77.3	70.2	40.6	30.8	26.8	23.2	18.6	17.0	17.
Total green weight, mg...	855	1080	1180	1250	1183	1133	775	425	319	245	165	150	133
Total dry weight, mg....	85	110	115	120	119	114	95	70	58	52	40	38	38

TABLE XIV  
 $\text{Fe}_2(\text{NO}_3)_6$  SERIES IN SOLUTION WITH NUTRIENT SOLUTION. OCTOBER 14—  
 NOVEMBER 1, 1905

	Nut. Sol.	$\frac{n}{7500}$	$\frac{n}{2500}$	$\frac{3n}{1000}$	$\frac{4n}{1000}$	$\frac{7n}{1000}$	$\frac{8n}{1000}$	$\frac{n}{100}$	$\frac{3n}{100}$	$\frac{5n}{100}$
Av. length of sprout, cm....	21.5	24.5	26.1	25.0	22.4	20.9	19.1	16.5	14.1	12.4
Total transpiration, gm...	30.6	33.7	37.2	35.6	33.3	31.5	25.8	22.6	21.8	19.6
Total green weight, mg...	1170	1500	1657	1380	1049	665	468	306	305	368
Total dry weight, mg....	109	120	126	112	102	94	81	73	72	70

## IRON CHLORID

Experiments with  $\text{Fe}_2\text{Cl}_6$  were carried out in order to compare with  $\text{Fe}_2(\text{NO}_3)_6$ . It was found that plants die in the same concentration, and growth is accelerated in equal proportion by the same concentrations in either solution, as is shown in the Table XV.

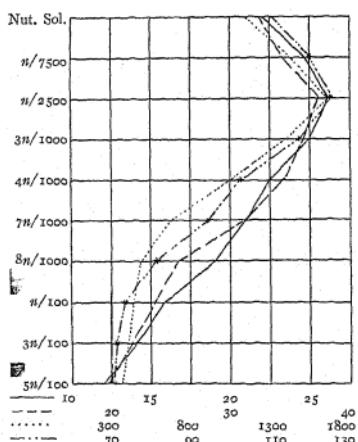


FIG. 28.—Iron nitrate series in nutritive solution. Oct. 14–Nov. 1, 1905.

TABLE XV  
 $\text{Fe}_2\text{Cl}_6$  SERIES IN SOIL. NOVEMBER 12–DECEMBER 12, 1905

	$\text{H}_2\text{O}$	$n/1000$	$2n/1000$	$3n/1000$	$4n/1000$	$6n/1000$	$7n/1000$	$8n/1000$	$9n/1000$	$n/100$	$2n/100$	$3n/100$	$4n/100$
Av. length of sprout, cm.	10.32	19.38	21.80	19.64	16.60	13.38	10.22	9.86	9.68	9.06	6.60	4.08	2.16
Total transpiration, gm.	85.30	75.80	78.10	62.9	41.10	36.40	27.70	29.20	29.10	29.10	30.70	24.00	24.10
Total green weight, mg.	787	845	835	652	490	450	235	205	188	119	40	35	26
Total dry weight, mg..	130	154	140	107	90	87	45	40	34	26	15	12	6

## NICKEL NITRATE

In nickel nitrate cultures is shown the most remarkable reduction of toxicity by the introduction of the quartz. Thus, in soil, plants die in concentrations  $7n/2000$  to  $6n/4000$ , while in solution death occurs at  $n/2000$  to  $n/4000$ . Stimulation effects are most marked in soil at  $n/2000$  to  $n/10000$ , but in solution at  $4n/10,000,000$  Tables

XVI and XVIII are the averages of three sets of experiments, the results of which are plotted in figs. 29, 30.

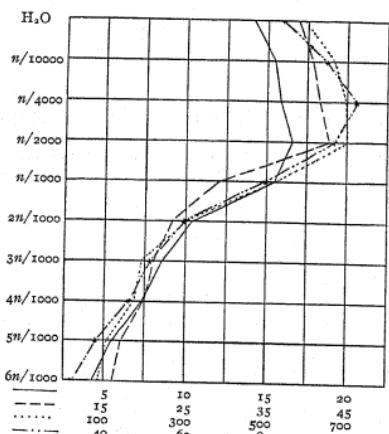


FIG. 29.—Nickel nitrate series in soil. Mar. 17-Apr. 2, 1906.

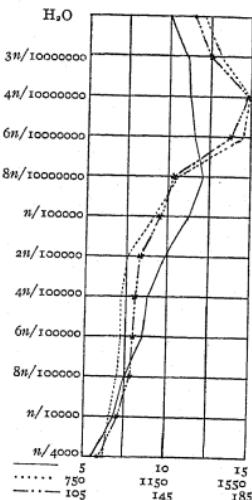


FIG. 30.—Nickel nitrate series in solution. Mar. 29-April 8, 1906.

TABLE XVI  
Ni(NO<sub>3</sub>)<sub>2</sub> SERIES IN SOIL. MARCH 17-APRIL 2, 1906

	H <sub>2</sub> O	$\frac{n}{10000}$	$\frac{n}{4000}$	$\frac{n}{2000}$	$\frac{n}{1000}$	$\frac{2n}{1000}$	$\frac{3n}{1000}$	$\frac{4n}{1000}$	$\frac{5n}{1000}$	$\frac{6n}{1000}$	$\frac{7n}{1000}$
Av. length of sprout, cm.....	14.16	15.40	15.94	16.50	15.20	10.30	8.50	7.30	5.40	4.30	Dead
Total transpiration, gm.....	39.3	41.7	42.8	43.6	29.4	23.1	21.0	19.7	17.4	17.3	
Total green weight, mg.....	574	685	696	668	518	200	185	160	103	91	
Total dry weight, mg.....	84	95	102	96	82	59	49	40	40	30	

TABLE XVII  
Ni(NO<sub>3</sub>)<sub>2</sub> SERIES IN SOLUTION. MARCH 29-APRIL 8, 1906

	H <sub>2</sub> O	$\frac{2n}{1000000}$	$\frac{4n}{1000000}$	$\frac{6n}{1000000}$	$\frac{8n}{1000000}$	$\frac{n}{100000}$
Av. length of sprout, cm.....	11.35	11.36	11.40	11.55	11.20	9.95
Total green weight, mg.....	1310	1345	1535	1490	1180	1000
Total dry weight, mg.....	154	160	185	170	140	140

TABLE XVII.—Continued

	$\frac{2n}{10000}$	$\frac{4n}{10000}$	$\frac{6n}{10000}$	$\frac{8n}{10000}$	$\frac{n}{1000}$	$\frac{n}{4000}$
Av. length of sprout, cm.....	8.66	8.21	7.40	7.38	6.83	5.50
Total green weight, mg.....	915	885	860	859	828	820
Total dry weight, mg.....	120	128	120	126	122	113

## PHENOL

In the phenol series death occurs at very nearly the same concentration in soil and in solution, viz.,  $2n/100$  to  $n/100$ . There is a very slight indication of reduction of the acceleration strength by the presence of quartz; thus, in soil the greatest growth occurs at  $6n/10000$ , in solution at  $4n/10000$ .

TABLE XVIII  
PHENOL SERIES IN SOIL. MARCH 17—APRIL 2, 1906

	H <sub>2</sub> O	$\frac{n}{1000}$	$\frac{2n}{10000}$	$\frac{4n}{10000}$	$\frac{6n}{10000}$	$\frac{8n}{10000}$	$\frac{n}{1000}$
Av. length of sprout, cm.....	12.52	13.94	13.56	14.50	13.98	13.70	14.48
Total transpiration, gm.....	30.1	43	44.0	44.9	48	43.7	40.1
Total green weight, mg.....	523	560	590	650	668	653	611
Total dry weight, mg.....	87	85	90	108	115	107	101

	$\frac{2n}{1000}$	$\frac{4n}{1000}$	$\frac{6n}{1000}$	$\frac{8n}{1000}$	$\frac{n}{100}$	$\frac{2n}{100}$
Av. length of sprout, cm.....	14.20	14.02	13.68	12.84	11.66	3.26
Total transpiration, gm.....	37.1	38.8	33.3	29.0	20.4	12.5
Total green weight, mg.....	523	518	488	470	403	68
Total dry weight, mg.....	95	85	77	72	54	17

TABLE XIX  
PHENOL SERIES IN SOLUTION. MARCH 8-22, 1906

	H <sub>2</sub> O	$\frac{n}{1000}$	$\frac{2n}{10000}$	$\frac{4n}{10000}$	$\frac{6n}{10000}$	$\frac{8n}{10000}$	$\frac{n}{1000}$
Av. length of sprout, cm.....	13.31	13.30	13.73	15.38	13.73	11.26	10.35
Total transpiration, gm.....	1148	1153	116	1101	1053	965	884
Total green weight, mg.....	140	142	146	148	140	131	122
Av. length of sprout, cm.....		$\frac{2n}{1000}$	$\frac{4n}{1000}$	$\frac{6n}{1000}$	$\frac{8n}{1000}$	$\frac{n}{100}$	$\frac{2n}{100}$
Total transpiration, gm.....		8.81	8.20	7.71	6.23	5.23	4.10
Total green weight, mg.....		823	701	640	575	521	440
Total dry weight, mg.....		114	103	101	83	82	70

These results are shown graphically in figs. 31, 32.

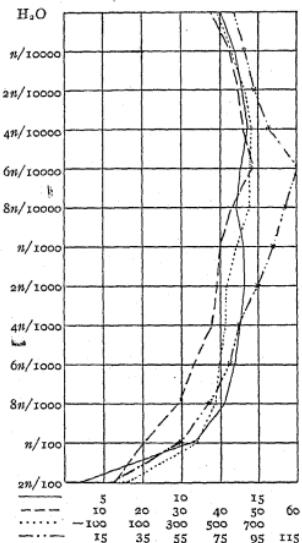


FIG. 31.—Phenol series in soil. Mar.  
17-Apr. 2, 1906.

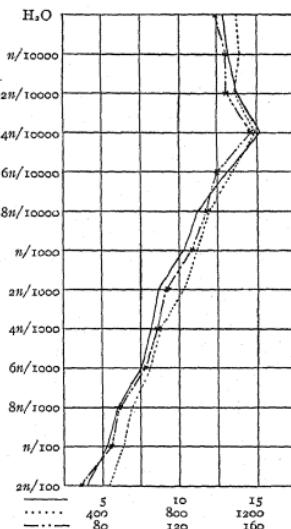


FIG. 32.—Phenol series in solution.  
Mar. 8-22, 1906.

#### ALCOHOL

Ethyl alcohol is less poisonous than anything thus far tried. Plants endure concentrations up to  $50n/100$  or  $75n/100$  and are most actively stimulated by  $75n/1000$  to  $25n/1000$  in both solution and soil. In the case of alcohol and phenol, therefore, the absorption of poison by quartz is practically a negligible quantity. Tables XX, XXI, and figs. 33, 34 show acceleration in soil at  $75n/1000$  and in solution at  $50n/1000$ .

TABLE XX  
ALCOHOL SERIES IN SOIL. MARCH 2-22. 1906.

	H <sub>2</sub> O	10n/1000	25n/1000	50n/1000	75n/1000	10n/100	25n/100	50n/100	75n/100	n
Av. length of sprout, cm....	19.02	18.92	19.34	19.02	20.64	18.64	15.46	11.14	6.16	3.44
Total transpiration, gm.....	38.80	37.80	37.90	38.70	40.30	31.30	24.00	14.50	13.70	6.90
Total dry weight, mg.....	600	640	649	661	705	611	510	310	80	50
Total green weight, mg.....	96	92	94	100	115	91	57	40	20	

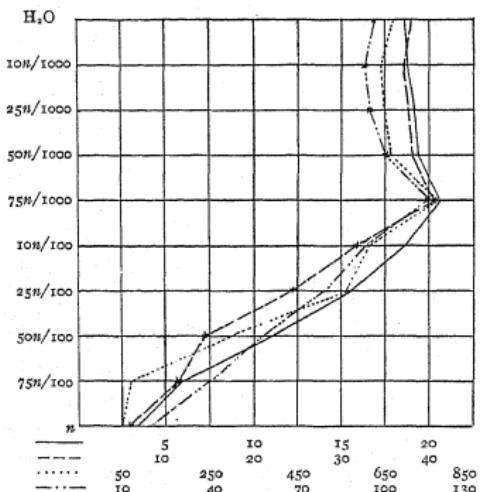


FIG. 33.—Alcohol series in soil. Mar. 2-22. 1906.

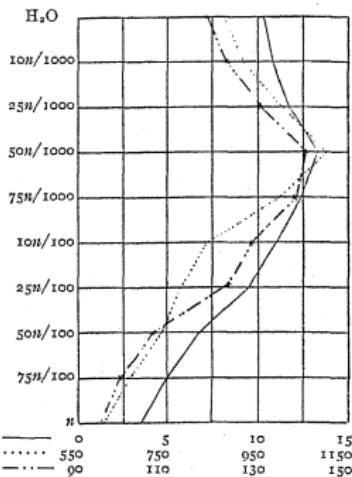


FIG. 34.—Alcohol series in solution. Mar. 2-22. 1906.

TABLE XXI  
ALCOHOL SERIES IN SOLUTION. MARCH 2-22. 1906

	H <sub>2</sub> O	10H 1000	25H 1000	50H 1000	75H 1000	10H 100	25H 100	50H 100	75H 100	n
Av. length of sprout, cm.....	10.30	10.85	11.83	13.35	12.23	10.96	9.43	6.66	4.81	3.40
Total transpiration, gm.....										
Total green weight, mg.....	877	910	1012	1109	997	831	778	729	653	513
Total dry weight, mg.....	115	122	130	140	138	126	127	100	97	95

## SUMMARY OF RESULTS

Table XXII shows the concentrations in which growth in quartz soil and in solution is stimulated and inhibited. From this it will be seen that in soil, nickel is by far the most toxic substance tried, being nearly five times as poisonous as zinc, silver, copper, six to eight times as poisonous as iron, and forty to one hundred times as poisonous as alcohol. On the other hand silver in solution cultures is more poisonous than nickel, zinc, or copper, the ratios being as 1 to 5, 7, and 10 respectively; while with iron, lead, and alcohol the ratios are 1 to 100, 400, and 7500.

It does not follow, therefore, that because a salt is highly toxic in solution it is equally so in soil; nor that one which holds a rela-

TABLE XXII  
SUMMATION OF KILLING AND STIMULATING CONCENTRATIONS SELECTED FROM FOREGOING DATA

POISON	SOIL CULTURES		SOLUTION CULTURES	
	Growth inhibited	Growth stimulated	Growth inhibited	Growth stimulated
Ni(NO <sub>3</sub> ) <sub>2</sub> .....	2n to 6n 1000	5n to n 10000	50n to 25n 100000	4n to 2n 10000000
ZnSO <sub>4</sub> .....	30n to 10n 1000	3n to n 10000	7n to 6n 100000	None
AgNO <sub>3</sub> .....	3n to 10n 1000	10n to 10n 10000	10n to 9n 100000	2n to 10n 10000000
CuSO <sub>4</sub> .....	3n to 10n 1000	10n to 4n 10000	100n to 50n 100000	None
Fe <sub>2</sub> (NO <sub>3</sub> ) <sub>3</sub> .....	6n to 40n 1000	9n to 30n 10000	100n to 900n 100000	4000n to 2000n 10000000
Fe <sub>2</sub> Cl <sub>6</sub> .....	6n to 40n 1000	9n to 20n 10000	100n to 800n 100000	4000n to 2000n 10000000
Pb(NO <sub>3</sub> ) <sub>2</sub> .....	50n to 30n 1000	9n to 40n 10000	4000n to 2000n 100000	20000n to 12000n 10000000
Phenol.....	20n to 10n 1000	8n to 4n 10000	2000n to 1000n 100000	800n to 4000n 10000000
Alcohol.....	750n to 250n 1000	750n to 250n 10000	7500n to 2500n 100000	7500n to 2500n 10000000

tively high position in the toxic table in soil should occupy the same relative position in solution cultures. Hence the assumption upon which HARTER (18) worked is not verified by my results.

The reduction of toxic effect (seen by comparing columns 1 and 3, Table XXII) is most marked in Ni, less in Ag, Zn, Cu, Fe, and Pb, in the order given, and no reduction whatever is apparent in phenol or alcohol.

Stimulation to growth takes place more markedly in soil than in solution, because in soil every test gave accelerating effects, while in solution cultures two failed to give such results. Stimulation in soil is always at a much higher concentration than stimulation in solution except in the case of the organic poisons tried, phenol and alcohol, where the presence of quartz does not reduce the toxicity.

### Conclusions

In conclusion I wish to point out what seem to me the most salient features of this research.

1. While considerable work has been done on toxicity, none has shown the definite effects of a poison in pure soil, for, as I have

elsewhere stated, such experiments have been made with garden loam and are unreliable because of the multiplicity of inorganic and organic substances with which the introduced poison may react. My results show that the introduction of pure quartz flour into a toxic solution, in such proportion as to form an ordinary moist soil, reduces the toxicity of the solution in a marked degree. But whether this reduction is due to adsorption, as TRUE and OGLEVEE would have it, or to a reduced freedom of movement of the solute particles, that is, a reduction of diffusion tension; as DANDENO suggests, or to some chemical changes enhanced by the quartz is yet an open question.

2. Of the limited number of poisons tried, all gave stimulation results in quartz, and all but two— $ZnSO_4$  and  $CuSO_4$ —in solution cultures. It is not beyond probability that these two, in proper concentration, might also be found to be stimulating.

3. The more dilute the fatal dose in solution, the more the toxic effect is reduced by the introduction of quartz. Hence the range of concentrations, both fatal and accelerating, is much greater in solution than in soil cultures.

Finally, I wish to acknowledge the friendly cooperation of all members of the botanical staff of the university, and especially the stimulating suggestions of Professor CHARLES R. BARNES and Dr. B. E. LIVINGSTON.

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## HYBRIDIZATION OF WILD PLANTS

D. T. MACDOUGAL

(WITH FOUR FIGURES)

THE number of forms of plants which have been or are regarded as hybrids by systematists is a large one and includes several oaks, of which two have been examined during the last two seasons. Attention has been called previously to the untrustworthiness of the custom prevalent among botanists of attributing a hybrid origin to certain plants because they appear to exhibit halved, fused characters, or a mosaic of qualities derived from the two supposititious ancestors. In some instances such deductions have been made by which the ancestry of a questionable plant has been made to include three or even four species. The argument of distribution is the main one offered in such attempted demonstrations. In many cases, this together with other circumstantial evidence may amount to almost positive conviction, but unless this close relation of well-joined facts is furnished, assertions as to the hybridity of a plant must be taken simply as a suggestion to be tested by cultural or experimental methods.

When confronted with an enigmatical plant of such character, three methods of attack are available to the investigator: that of obtaining the supposed hybrid by synthetizing it from its supposed parents; that of making an anatomical examination of the hybrid and the parents to which it has been referred; and that of obtaining second, third, and succeeding generations of the hybrid for the purpose of ascertaining whether or not any separation of the ancestral characters may occur in an alternative inheritance by which the ancestral forms may actually reappear. All of these methods are beset with numerous difficulties, but when used together with the facts of distribution a very satisfactory degree of proof may be obtained. It will be profitable to consider the scope and application of the various methods of experimentation and observation noted above.

\* Read before Sections F and G, A. A. A. S., New York City, December 27, 1906.

The re-formation of a hybrid by the cross-pollination of the parents to which it may be ascribed is by no means simple in all instances, nor is it always easy of accomplishment. In the first place, the original cross-pollination may have taken place possibly under an exceedingly rare combination of favorable physiological conditions difficult to secure or duplicate in experimentation. Then one or both of the species as ordinarily recognized may in reality consist of two or more elementary species, which may not differ widely in external anatomical characteristics, but exhibit widely divergent physiological characters and behave quite differently in breeding. A hybrid with one of these forms may differ in very many important particulars from a hybrid with another constituent of the same species. Of course if we deal with elementary species only in our usage of the term, this difficulty does not exist; but it does appear as a serious matter with the customary practice, as has been found in a number of breeding experiments, and furthermore must be taken into account no matter by what method we consider a hybrid.

Let us suppose, however, that we have actually in hand the two strains or elementary species by which the hybrid may have arisen, and we have still one more matter which may mislead us. This consists in the fact that reciprocal crosses are not always identical in their products. Thus the pollen of *A* and the egg of *B* do not necessarily make the same hybrid as the egg of *A* and the pollen of *B*.

The pollination of *Oenothera Lamarckiana* by *O. biennis* generally results in securing a progeny separable into seven types, some of which are stable and reproduce themselves exactly in succeeding generations, while others split into two or more forms in the second generation. On the other hand, the use of pollen of *O. Lamarckiana* on pistils of *O. biennis* results in a progeny embracing four types, none of which is identical with the components of the reciprocal cross. *O. biennis* shows a similar behavior in some other crosses in the limited observations recorded. In all such cases it must be understood that the number of types does not appear to be invariable, and that a progeny of a hundred thousand is likely to include more than one of a score. It is evident that in the determination of a hybrid by this method difficulties may be met with. Thus the failure of the operator to secure the supposed hybrid may not be considered

as final proof that it may really have come by one of the rarer combinations which he has missed. On the other hand, success may come with the first cross and in the first generation.

In some instances the result of a hybridization is a single type which offers the qualities of the parents locked in a stable combination in the first generation and reproducing without separation in successive generations. It is this type of hybridization that is implied in the general assertions as to the hybrid origin of any plant, and it is a type of which we have the fewest illustrations in breeding experiments. To recur again to the genus furnishing the example previously given, *O. cruciata varia* was suspected by the author to be a combination of *O. Lamarckiana* and *O. cruciata*, and in the synthetization test the good fortune was encountered of selecting the one of the three known elementary species of *O. cruciata* which had originally entered into the union. The egg of *O. Lamarckiana* and the pollen elements of this form entered into a stable combination which has the distinctness and fixity of a species, and as a matter of fact this hybrid has been long mistaken for the true *O. cruciata* by a great number of European gardeners and botanists.

In addition to the difficulties of hybridization and interpretation of the results described above, it is also to be taken into account that in some instances a long period ensues between the act of pollination and the perfection of the fruit, and then a long time is necessary for the germination of the seeds and development of the progeny. Ten, fifteen, or even twenty years might be necessary to make an application of this method to some of the species of trees, which would obviously make it unavailable except under extraordinary circumstances.

The examination of the anatomical characters of a plant to determine its ancestry is a method which has become of less esteem in the light of modern additions to information as to the character and behavior of hybrids. Of the various types of hybrids described it is of course the fixed hybrid which is most likely to come under examination, and while it occasionally presents a fair average of the characters of the parent, more frequently it is gonoecolic to one or the other, and may be so near one parent that a gross or minute estimate of the tissue structure would offer nothing better than a guess as to the

other parent, a guess which might be fortified to some extent, perhaps, by the facts of distribution. In certain meristic qualities, such as the rate and total amount of growth, the hybrid may even exceed that of either parent, or be less than either.

The mistaken impression prevails that hybrids bear defective pollen, but this is generally found to be the case only when the cross is unbalanced in many characters, particularly those appertaining to the reproductive functions. The infertility of many animal hybrids has also strengthened the assumption that plant hybrids share this defect. Many hybrids are quite as prolific in mature seeds as either parent, while in some that are offered by the seedsman and nurseryman they are claimed to excel in this respect.

In any case where the comparative anatomical method is used, care must be taken to make observations upon material from similar stages of development. This is forcibly impressed upon one after following the growth of a hybrid which in the seedling stages shows a predominance of the qualities and anatomical characters of one parent, a different arrangement in the adult shoot, and a still different balance in the flower and fruit.

The third method of study of a supposed hybrid is one which involves pure cultures of its progeny for one or two generations. If it should be a fixed hybrid no results will be secured which will be of value in the solution of the problem, since, so far as any facts offered by such cultures are concerned, the plant behaves as any other species. Presumably most of the species of suspected hybrid origin are of this character, but some of them undoubtedly will be found to be constantly re-formed and to offer alternative inheritance, and hence this test should be applied whenever practicable.

If the supposed parents differ in but one or a few characters, and the hybrid shows alternative inheritance, the solution of the main question lies near at hand. It is not such simple questions as this however that we are usually called upon to solve. The real difficulties lie in the hybrids with the component qualities in stable combination, making a fixed hybrid, and with the forms which exhibit an interlocked combination of the ancestral characters in the first generation which resolve into the possible combinations of the dissimilar characters in the second, by which an enormous range of indi-

viduals is presented, which without close examination of a large progeny might appear to be a hopeless medley. This is best illustrated by a plant which has been studied recently in cultures at the

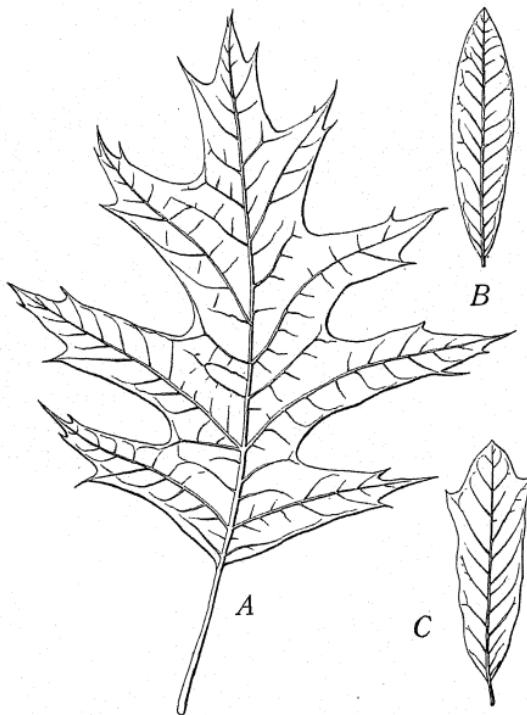


FIG. 1.—*A*, leaf of *Quercus rubra* from N. Y. Botanical Garden; *B*, leaf of *Q. Phellos* collected at Ashland, N. J., Sept. 14, 1892; *C*, leaf of *Q. heterophylla* taken from tree from which acorns were taken for cultures, collected at Richmond Valley, Staten Island, N. Y., October 22, 1905.— $\times \frac{1}{2}$ .

New York Botanical Garden, and at the Desert Laboratory, Bartram's oak, *Quercus heterophylla*.

Bartram's oak was discovered as a single individual growing on a farm of Mr. John Bartram, near Philadelphia, on the banks of

the Schuylkill at some time previous to 1750. A rather complete history of the occurrence of other individuals which were included with this tree under the name of *Q. heterophylla* is given by Dr. ARTHUR HOLLIICK in *Bull. Torr. Bot. Club* (15:303. 1888) and need not be repeated here.

After a consideration of various lots of material which appeared to differ widely, various botanists have regarded this tree as a hybrid between *Q. Phellos* and *Q. tinctoria*, *Q. Phellos* and *Q. falcata*, *Q. Phellos* and *Q. coccinea*, *Q. Phellos* and *Q. palustris*; while others have designated it as a variety of *Q. Phellos*, *Q. coccinea*, *Q. aquatica*, *Q. nigra*, and *Q. imbricaria*. It is notable that of the botanists who ascribed a hybrid origin to the plant all agreed that *Q. Phellos* must be one of the parents, a fact which will be easily explainable when an examination of its leaves is made.

In October 1905 the author was accompanied to a locality on Staten Island by Dr. HOLLIICK and Dr. BRITTON, where several trees of the species had been under observation by them for many years. About 75 acorns were procured from a tree which bore leaves of a form approximating that of figs. 2 and 3, and were placed in the propagating houses of the New York Botanical Garden, with the result that 55 plantlets were available for study in December and January following. With the formation of the earliest leaves it became evident that a wide diversity of form of these organs and of other qualities prevailed, as shown by the photograph taken in April.

In May 1906 all of the plantlets were transferred to the experimental grounds, and as development proceeded the diversity became still more marked. At the close of the season it could be seen that this group of plants included some which simulated *Q. Phellos* with its lanceolate entire leaves, while others were not separable from *Q. rubra*, the remainder being capable of arrangement in a series between these two poles. An examination of the literature disclosed the fact that the combined observations of the several botanists who have written on the subject refer to plants bearing almost the entire range of leaves noted in the cultures here described. In most of these accounts the leaves are said to be much like those of *Q. Phellos*, while some observations include notices of others which were broad lobed and notched, although most of these writers were extremely chary of

identifying any of the forms with those of *Q. rubra*. It is to be noted, however, that as a result of the consideration of gross anatomical facts and distributional data, Dr. HOLICK and other botanists had finally concurred in the general conclusion that the tree was in all probability a hybrid between the red oak and the willow oak.

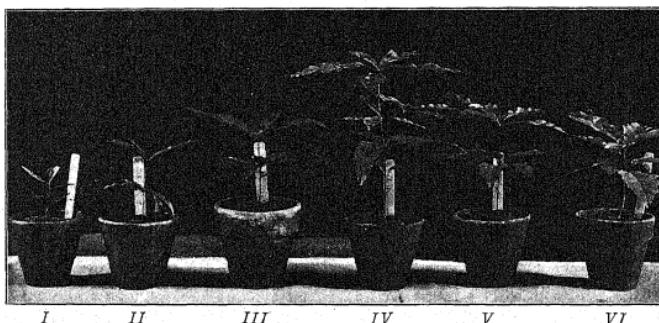


FIG. 2.—Plantlets from progeny of tree of *Q. heterophylla* which bore leaves as in fig. 1, C. I bears leaves not distinguishable from those of *Q. Phellos*, and VI resembles *Q. rubra*. II, III, IV, and V form a series between I and VI.—Photographed April 1906.

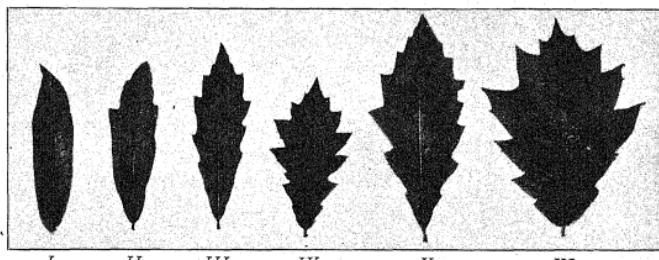


FIG. 3.—Series of leaves from progeny of *Q. heterophylla*, parallel to those shown in fig. 2, but taken at the close of the season of 1906.

With our present available information concerning the behavior of hybrids this conclusion seems unavoidable. If we attempt to follow out the history of the hybridization as it may have occurred, however, we are compelled to rely upon inference in part. The

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actual nature of the immediate product of the cross is not known. The parental species have undoubtedly sustained similar distributional relations to each other for uncounted hundreds or thousands of years, and there is no reason to suppose that hybridization may not have taken place many generations ago. On this account it is not possible to say whether the tree from which the germinated acorns were taken was the immediate product of the cross or whether it is the  $n$ th generation of its progeny.

In balanced crosses in which the parents show a large number of dissimilar characters, the first generation rarely offers the spectacle of pure dominance of the characters derived from one parent, and recessiveness of those from the other parent. It is only when the parents differ by a point or two only that such total dominance is seen, and the first generation or the immediate product of the cross resembles one parent or the other, and its progeny split in the next generation. In cases such as that under discussion, and which is also illustrated by the walnuts, the first generation shows a mixed dominance as well as a possible fractionization of some meristic qualities, so that the hybrid appears as an intermediate between the two parents, toward both of which its relative position may be variously estimated. In the second generation the movements of the recessives coupled with the range of fluctuating variability should give a wide diversity of types, varying in number with the number of differentiating points of the parents of the cross, which may include both parents, the type of the first generation, and an intricately interwoven connecting series of forms.

By reason of the number of dissimilar characters involved in such a cross, the probability of deriving an individual composed entirely of recessive characters, or of the particular combination characteristic of either ancestor is very small. A progeny of hundreds of thousands of species would be necessary to furnish a series inclusive of both ancestors and intermediate combinations.

While it may not be said that any of the plantlets of the progeny under observation are reconstituted ancestral forms, yet some of the individuals include so many of the qualities of the red oak and willow oak that the evidence is overwhelmingly in favor of the conclusion that the origin of *Q. heterophylla* is to be attributed to the hybridiza-

tion of these two forms. Taking this conclusion as established, it may then be said that the name *Q. heterophylla* is at present applied to a medley of oak trees which possibly includes the first generation of a cross between *Q. rubra* and *Q. Phellos*, secondary hybrids with either parent, as well as successive generations in which various combinations of ancestral qualities may appear.

Another aspect of the parental form of the above hybrid and the progeny remains to be mentioned. A collector covering the field occupied by the hybrid in which the parental forms come into contact, who gathered a full series of material from the trees available, would have data upon which mistaken conclusions as to intergradation of species by fluctuating variability might be made. This leads to the suggestion that any supposed intergradation of two species of seed-plants should be examined with respect to possible hybridizations before any final estimate is reached in the matter.

Furthermore, it is to be seen that while in all reasonable probability opportunity for hybridization between these two oaks has been present for a period of unknown but undoubtedly great length, it has not resulted in anything in the way of occurrence or distribution suggestive of the disappearance of either parental form. The probably greater frequency of intra-specific fertilization over hybridization would secure this result. Then again it is to be seen that even in the case of complete cross-fertilization of all of the individuals there would be the probable reconstruction of the ancestral forms among the progenies.

*Quercus Rudkinii* has long been reputed to be a hybrid between *Q. Phellos* and *Q. marylandica*, and a visit was made to the group of trees from which the species was originally described by Dr. BRITTON, in company with him and other botanists in October 1905. These trees stand near Cliffwood, New Jersey, and since the original discovery in 1881 others have been found on Staten Island and also to some distance to the southwestward in New Jersey. The mere facts of distributional relations together with the anatomical features offered by the bark and leaves led to the description of these trees as being of a hybrid origin from the willow oak and the black oak. If these facts only are taken into account, it seems quite as plausible to regard this tree as a hybrid as *Q. heterophylla*. The leaves from the included trees showed a range of forms that included the type

of *Q. Phellos*, but did not go wide enough to show duplicates of those of the other parent as described and figured by Dr. BRITTON (*Bull. Torr. Bot. Club* 9:13. 1882). The acorns likewise ranged from the form near that of *Q. Phellos* toward *Q. marylandica*, but included none that might be mistaken for the latter. The bark of the various individuals, on the other hand, seemed most like *Q. marylandica*, although much variation was apparent (fig. 4).

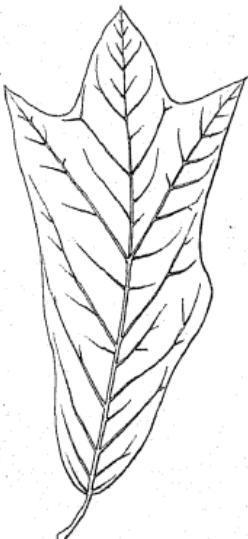
During the visit of 1905 acorns were found on trees of some of the types only, and material capable of germination was obtained from three of them. When these were at the end of the first year's growth, which began in the propagating houses in January 1906 and ended in September, it was seen that the progenies of the three parents, selected for their dissimilarity, were fairly identical; and while a wide range of variability was found, this range did not exceed the limits of similar fluctuations offered by other species of oaks which are known to be unified hereditary strains.

Over a hundred plantlets were carried through the first season, but no leaf-forms were exhibited which might be taken to simulate either of the supposed ancestors, although a comparative culture of seedlings of these might have brought some resemblances to light. The advanced stages,

FIG. 4.—Leaf of *Q. Rudkinii* taken from tree at Cliffwood, N. J., from which acorns were taken for cultures.— $\times \frac{1}{2}$ .

however, may modify this aspect of the progeny materially. Some of the trees examined bore only imperfect acorns, and the proportions incapable of germination taken from all of the individuals was large. This and other facts suggests a more widely unbalanced cross than in the case of Bartram's oak, and it is quite possible that the dissimilarity may be so great that a finely graded series or a total series may be impossible in the actual combinations effected.

As the facts now stand, however, the observations have not given any actual proof of a hybridization, although in view of the above



considerations this is not disproved. Synthetic tests, which would consume a large fraction of a human life-time, and anatomical examinations are yet to be resorted to. Meanwhile this oak, which seems to be constant in successive generations within the limits of its variability, must be regarded as a species in accordance with current taxonomic practice, until some positive evidence to the contrary is obtained.

The principles illustrated by the foregoing facts may be briefly summarized as follows:

It is obvious that the facts of geographical distribution may be relied upon to furnish conclusive evidence as to the origin of a species or a hereditary quality only under very exceptional conditions, in which other possibilities are excluded, and then only in a circumstantial manner. It is of course a basal and necessary fact that species not in contact may not hybridize, but the converse is true only when otherwise proven.

So far as the plants of suspected hybrid origin from parents suggested by distributional relations are concerned, the methods of investigation available are two, which may separately secure affirmative evidence of conclusive value, while the third may bring no more than confirmations and suggestions.

Attempts at synthetization, if successful, yield dependable conclusions as to the composition of a hybrid, yet a failure to secure a form by synthesis may be due to innate and almost intangible difficulties in the hybridization of the forms concerned, by the different results of reciprocal crossing, and the difference in physiological attributes of elementary species included under one name. Furthermore, the natural form, the ancestry of which is under search, may have been a derived hybrid which became fixed in the  $n$ th generation by a fortuitous combination of dominant characters. To secure a similar result in an experimental test might be beyond the range of probability.

In an anatomical examination such a combination of dominant and recessive characters with fluctuations in meristic features away beyond either parent may make the results of but little value until confirmed by data derived from other sources, before their full value may be known.

A study of a fixed hybrid by cultural tests of its progeny will reveal

nothing as to its origin, and synthetization and anatomic examinations are the only recourse. On the other hand, if the progeny exhibits alternate inheritance, its components may show unmistakably the nature of the original cross.

The last-named method demonstrates beyond reasonable doubt that Bartram's oak is a hybrid derivative of the willow oak and red oak, the progeny being probably characterized by alternative inheritance of some of the qualities, and fractionization of others, instead of being a unified hereditary strain. Some of the plantlets included in a progeny of 55 individuals were apparent re-constructions of the ancestral types as observed at the close of the first year, while others were variously intermediate.

Rudkin's oak, on the contrary, yields no evidence in cultures of its progeny on which a defensible conclusion as to its origin may be based. The individual variability of the trees included under this name is very great, and some of these approach the willow oak in leaf-form, and some in the character of the acorns. The study of a large number of seedlings from three trees showed the willow oak in leaf-form and some of the characters of the acorns. The study of over a hundred plantlets showed a wide and practically identical range of variation. Alternative inheritance could not be traced. The individuals known under the name of *Q. Rudkini* produce a large proportion of imperfect acorns, but beyond this no facts suggestive of hybrid origin can be found, except the anatomical resemblances noted.

A list of the natural plant hybrids of North America was prepared by Mr. DAVID GEORGE for presentation at the International Hybrid Conference in New York in 1902, but was not published. In this list it was noted that 117 natural hybrids had been reported as occurring in the indigenous flora of North America, in addition to some instances among the ferns and mosses. The manuscript having been placed at my disposal by the director of the New York Botanical Garden, a revision of the list has been made, some of the original references being omitted and a few added. No attempt has been made to make it actually complete, the sole purpose being to suggest material for extended observations similar to those described in this paper. The reported hybrids are distributed among the natural families as follows:

NAIADACEAE.—Two cases in *Potamogeton*.

CYPERACEAE.—Twelve hybrids in *Carex*.

JUNCACEAE.—One hybrid between *Juncus effusus* and *J. pacificus*.

LILIACEAE.—One hybrid between *Calochortus Benthami* and *C. albus*.

ORCHIDACEAE.—One hybrid between *Habenaria lacera* and *H. psycodes*.

JUGLANDACEAE.—Three hybrids of *Hicoria pecan* with *H. minima*, *H. alba*, and *H. laciniosa*; one hybrid between *Juglans nigra* and *J. cinerea*.

SALICACEAE.—About twenty supposed hybrids of *Salix* are noted; two hybrids of *Betula* are also reported, viz., between *B. pumila* and *B. lenta*, and between *B. populifolia* and *B. papyrifera*.

FAGACEAE.—Thirty-five oak hybrids have been reported, but one of these, *Q. Rudkini*, has been tested, as described above, with negative results.

RANUNCULACEAE.—A hybrid between *Actaea alba* and *A. rubra*; *Clematis viornaoides* is reported to be of a hybrid nature.

CRUCIFERAE.—*Roripa palustris* is supposed to form natural hybrids with *R. obtusa* and *R. sinuata*.

POMACEAE.—*Malus Soulardii* is taken to be a cross between *M. coronarius* and *M. Malus* by some workers.

ROSACEAE.—*Geum strictum* and *G. canadense* are supposed to form a hybrid.

PAPILIONACEAE.—A hybrid between *Baptisia australis* and *B. bracteata* is reported.

ANACARDIACEAE.—*Rhus hirta* is supposed to cross with *R. glabra*.

RHAMNACEAE.—*Ceanothus Lobbianus* is taken to be a hybrid between *C. thyrsiflorus* and *C. dentatus*; it is also supposed that *C. thyrsiflorus* hybridizes with *C. papillosus* and *C. sordidus*.

VIOLACEAE.—A large number of hybrids of *Viola* have been reported.

ONAGRACEAE.—*Oenothera biennis* and *O. Oakesiana* hybridize, the progeny of the first generation consisting of several types.

CACTACEAE.—A hybrid is supposed to be formed between *Opuntia fulgida* and *O. spinosior*.

CORNACEAE.—*Cornus Baileyi* has been taken to be a hybrid between *C. asperifolia* and *C. stolonifera*.

VERBENACEAE.—The evidence seems strong that *Verbena canadensis* and *V. bracteosa*, *V. bracteosa* and *V. hastata*, *V. bracteosa* and *V. urticifolia*, *V. bracteosa* and *V. stricta*, *V. stricta* and *V. hastata*, *V. stricta* and *V. urticifolia*, *V. urticifolia* and *V. hastata*, *V. angustifolia* and *V. stricta*, *V. stricta*, *V. angustifolia* and *V. bracteosa* hybridize.

ACANTHACEAE.—*Ruellia ciliosa parviflora* has been supposed to be the result of a cross between *R. ciliosa* and *R. streptocarpa*.

LOBELIACEAE.—*Lobelia syphilitica* is supposed to hybridize with *L. cardinalis*.

CICHORIACEAE.—*Prenanthes Mainensis* is taken to be a cross between *Nabalus racemosus* and *N. trifoliatus*.

COMPOSITAE.—Hybrids are reported in *Eupatorium*, *Solidago*, *Aster*, *Bidens*, and *Helenium*.

The genera noted above are supposed to offer about two hundred hybrids, and as the observations have been made principally with the flora of eastern North America, in a region which probably does not furnish more than four thousand species, it is to be seen that the questions involved affect about 0.5 per cent. of the flora. Probably not more than half of the instances included in the above list could be confirmed by actual tests; but on the other hand a closer examination of other genera would reveal an equal number of actual occurrences.

Some of the constituents of the native flora are known to be constant unitypic hybrids, and hence have every claim to be regarded as species. The type of hybridization most widely different from this is illustrated by Bartram's oak, and between these two diverse modes of action may be found. In gaining a more extended and accurate knowledge of the manner in which the qualities of separate unified strains of plants are alternative, or are interlocked, or fractionized in hybridizations, a vantage ground will be gained for the consideration of all questions in genetics as to saltations, minute accretions, and fluctuations of such characters.

DEPARTMENT OF BOTANICAL RESEARCH,  
CARNEGIE INSTITUTION OF WASHINGTON.

## BRIEFER ARTICLES

### ILLUSTRATING BOTANICAL PAPERS

Long editorial experience in various methods of illustration of the *BOTANICAL GAZETTE* has convinced me that much more adequate and satisfactory results might be secured if authors knew something of the modern methods for reproducing photographs and drawings, and particularly of the requisites for success. Requests for information of this kind, as well as the need felt in the editorial rooms, have determined me to discuss, as briefly as possible, the more important points.

The first question for author and editor to settle is this: Shall the figures be distributed in the text or aggregated into plates? Too often this is unconsidered or decided in conformity to a custom which arose out of necessity in the past. The process of engraving wood and metal by hand was the original and very costly method of illustrating books and monographs. (The journal and short paper were not then in existence.) The invention of lithography offered a vastly cheaper method, which was quickly adopted. The advantage of having figures close to the text they illustrated was surrendered, chiefly on account of the financial advantage, and partly because better effects could be secured by the new process. Modern methods, however, have made possible again the use of the text cut at the point where the figure will be of the most service to the reader. It is highly illogical, therefore, to conclude that because lithographed plates were used in the last century to illustrate the best scientific treatises and monographs, this mode of illustration is today evidence of first-class work. Yet novices are liable to precisely that misconception. There are some cases in which plates are still preferable to text cuts; e. g., when a large series of figures must be before the eye at once, or when the same figure must be referred to at many points. But these cases are much rarer than the prevalence of plates would indicate. Rather this prevalence indicates a want of consideration by the author, who from habit demands plates for a paper, but would never think of illustrating a book so. Having determined whether text cuts or plates are best, the mode of reproduction must be selected, for it is absolutely necessary to adapt the drawings to the chosen mode.

At present the following forms of illustrations appear in scientific journals: (1) lithographs; (2) photolithographs; (3) photogravures; (4)

half-tones; (5) copper and zinc etchings; (6) wood-cuts. I propose to state the nature of each of these, its limitations, its adaptability to special needs, and the requisites, so far as the author is concerned, for successful reproduction of illustrations by each process. Some examples of each are cited from the recent volumes of the *GAZETTE*. This citation does not imply that the "copy" was what it should have been; but when it was not the citation is made in order to show what can be done even when the drawing was poorly adapted to a particular mode of reproduction.

1. **Lithographs.**—These are made by drawing with crayon or pen on a stone or metal plate the design to be printed. It requires an expert draftsman, accustomed to botanical work, to make such drawings well. Even at best the draftsman may make mistakes, or introduce interpretations foreign to the author's design. The chances of error increase the poorer the original drawing and the more the lithographer has to alter it to make it presentable. Thus the only advantage of the intervention of an artistic hand is accompanied by a distinct disadvantage, and this disadvantage is increased by the likelihood that he who can only make a poor representation of his observations is the more likely to overlook errors on the part of the draftsman.

In this country the cost of lithography is almost prohibitive, and the work on histological and cytological subjects is rarely good. The inevitable delays, if drawings are sent to Germany and proofs returned, not to mention other difficulties, make it almost impracticable to attempt lithography. And if good drawings can be furnished, other methods are far better, because they provide for photographically exact reproduction. The only possible reason why a lithograph should be desired lies in the necessity of making poor drawings presentable. This should be obviated, if a second hand must intervene, by employing an artist to make drawings which can be reproduced photographically. Money is much more wisely spent on clear and accurate drawings, which can be corrected at leisure, than on expensive lithographs, with the artist across an ocean or a continent.

Examples:—32: pls. 1-3 (German work, cytological); 33: pls. 10, 11 (American work, entire plants); 30: pl. 12 (American work, cytological).

2. **Photolithographs.**—These are made by covering the surface of stone or metal with a composition which will become insoluble when exposed to light and printing on this prepared surface a photographic positive of the design. After further preparation the design may be printed on paper by inking the face of the plate. Such printing is done by special power- or hand-presses and cannot accompany the text. It

is available therefore for plates only. It is not an expensive process and gives better results than zinc etchings. It is recommended whenever plates are required. The drawings must be of the same kind as for copper and zinc etchings (see 5).

Examples:—**35**: pls. 4, 5; **36**: pls. 17-20 (printed in paler tinted inks); **37**: pls. 14, 15, 26, 27; **38**: pls. 1-4; and especially **42**: pls. 19-28.

**3. Photogravures.**—These are printed from an etched metal plate or from relief plates, the design being reproduced by rather elaborate mechanical and photographic means. The process is unsuited to anything but plates; and as the prints must be made separately on hand-presses, photogravure is expensive, costing three to four times as much as photolithographs, zinc or copper etchings, or half-tone work. The advantage is that photographs and all kinds of drawings (whether in ink, crayon, or pencil; whether in line or tint or wash) can be reproduced with the utmost fidelity, with an almost perfect rendering of tone and of light and shade. Pencil drawings, however, may be too weak in contrast to permit effective reproduction; and inasmuch as contrast is always somewhat reduced, shaded drawings should show a little more than is desired.

Examples:—**33**: pl. 1 (from pencil drawings); **34**: pl. 17 (from pen and wash); **37**: pls. 11-13 (from pencil and wash); **38**: pls. 6, 7 (from pencil, background removed).

**4. Half-tones.**—These are relief plates, usually of copper, in which the "grain," instead of being of almost imperceptible dots irregularly distributed as in photogravure, is produced by a screen of ruled lines, whose intersections form equally spaced dots, isolated, or blended with others into irregular lines or surfaces. Half-tone work is adapted to photographs and to varied styles of drawing, like photogravure; but as the "grain" is coarser it does not equal that process in delicacy or range. Yet when the screen used is fine and the workmanship first class, the results are almost as good. It has the very decided advantage that the metal plate may be mounted on a block and may then be printed along with type. It is necessary, however, to use a very smooth paper, either highly calendered or coated with clay. The coated papers are believed to be not durable and are to be avoided whenever possible.

Half-tone work alone is available for reproducing photographs to be printed in the text. Good prints (not negatives) are needed, showing sharply the details desired. Almost any sort of print can be reproduced; but engravers prefer, doubtless from habit, those made on Solio paper, toned brown. In case any painting-out of defects or background is to be done, unglazed prints are preferable.

If wash or pencil drawings are to be reproduced, contrast should be greater than desired, since there is necessarily a light gray background representing the whites of the picture. Except when figures have too intricate outlines this background can be cut away.

Examples:—35: 198-207; 38: 48-57; 37: 304-5 (which shows what can be done with simple apparatus by cutting away and vignetting the background); 39: pl. 8 (pencil drawing, background cut away); 39: pl. 4 (2d issue; from water color); 41: pls. 1, 2 (from photomicrographs; compare wth photogravure from similar originals, 32: pls. 15-17); 41: pls. 3, 4 (from pen and pencil, line and stipple, combined with wash; compare with potogravure from similar original, 37: pls. 11-13).

5. **Copper and zinc etchings.**—All the previous processes are as truly dependent on etching by some solvent as are these. The term is used to distinguish the copper or zinc plates, made without grain or screen, mounted type high and intended for printing on an ordinary press. Copper is used for the finer and more delicate work; zinc where the work is bold. Naturally the copper block is the more expensive. This process is in all ways the one best suited to reproduce drawings for text use. It is also satisfactory for plates; indeed there is scarcely any work that cannot be adequately and conveniently illustrated by it, if only the drawings are made with reference to this mode of reproduction.

Drawings must be in dead black ink (BOURGEOIS AINÉ, Encre de Chine Liquide; WINSOR and NEWTON, Liquid India Ink; HIGGINS, Waterproof Drawing Ink), and always in line and stipple only. Pale ink or a wash or tint make the use of the process impossible, unless the etching is supplemented by a second block in half-tone. Drawings should not be less than one half larger than the reproduction is to appear; preferably they should be of double size, and for open diagrams or charts treble or even quadruple size may be better. This permits refinement of line and fine shading without excessive care in making the original. To see how a coarse drawing will look a concave lens 4 or 5<sup>cm</sup> in diameter should be available. There are few observers who can draw at all, and none who draw well with a pencil, who cannot readily acquire the art of drawing with ink in a style suitable to be reproduced by copper or zinc etching. This gives freedom to put illustrations in the text where this is desirable, or to combine them into a plate if necessary.

Examples:—26: pls. 1, 2 (a fine example of pure line); 32: pls. 10-12 (almost pure stipple); 33: pl. 14 (line and stipple); 36: facing p. 188 (map, letters and figures pasted on; red index lines and figures by second printing); 39: facing p. 102 (map, two blocks made from single drawing

and printed separately); **38**: 347-361 (graphs and intricate tabular work; note especially insert after p. 356); **40**: pls. 10, 11 (line with wash; block a combination of half-tone and zinc; not recommended).

**6. Wood-cuts.**—The expense of this process is prohibitive, especially in view of the many mechanical processes whose results are equal or better.

**General suggestions.**—It is advisable for convenience in sending drawings and photographs by mail that they be unmounted. If they are to be arranged in groups that cannot be designated by serial numbers alone, they may be arranged as desired on a sheet of thin paper, the outline of each roughly traced thereon, and the chart folded and forwarded with the separate sketches or photos in a small package.

Index letters should be printed and pasted on at the end of index lines. Avoid the use of more than one letter at each line, but keep the same letter for the same structure throughout. *Syn* is no more significant, as applied to synergids, for anyone competent to read the paper, than the letter *s* or *x*. Sheets of printed letters and numbers will be supplied on request.

It is very instructive to compare original drawings and reproductions; much can be learned from a single experience which can be applied to modifying future work.

For drawing, an ordinary sharp Spencerian or Gillott pen is better than the excessively fine lithographic and crow-quill pens. Fine lines and very fine dots are not only entirely unnecessary, but too often they disappear in etching the plate, when moderately coarse lines and dots are properly reduced. Bold drawing on a large scale with proper reduction gives better results than the fine work done under a lens on a scale that permits only one fourth reduction or less. Plates may be printed in paler inks or tints to secure softness. Drawings unsuited to photographic reproduction are often excused on the ground that the author drew indistinctly because he saw the outline faintly. But a drawing which is dead black and shows the harshest contrast in order to be reproduced photographically, may be printed in as faint a tint as desired, thus reducing contrasts and eliminating the objectionable harshness. Where extreme gradations of tone are necessary, of course half-tone or photogravure should be employed.—  
CHARLES REID BARNES, *The University of Chicago.*

## CURRENT LITERATURE

### BOOK REVIEWS

#### Some elementary textbooks

The series of well-known and very successful elementary texts of BERGEN has been increased by a new one in collaboration with DAVIS.<sup>1</sup> Every successful teacher of botany recognizes that there are possible different lines of approach to the subject. Hence it is quite usual for textbooks to be so arranged as to permit selection and rejection to fit the desires of various teachers. However, when two plans of organization are carried out practically throughout the whole plant kingdom, really two courses in botany are provided for. Such is the arrangement of the book under consideration, which consists essentially of two books bound together.

Part I, on "The structure and physiology of seed plants," and Part III, on "Ecology and economic botany," comprising 299 pages of the book, are by Professor BERGEN. They have a distinctive method of presentation, and include most of the topics found in the widely used and successful *Foundations of botany* which the present book is designed to supplant. Part II (256 pages), on "The morphology, evolution, and classification of plants," is by Dr. DAVIS. Parts I and III are topical in nature, while Part II is arranged according to the increasing complexity of the plant kingdom. The authors suggest that "the whole will furnish material for a full year's work," usually omitting portions, and "that a half-year course can be readily arranged by selections from the more general sections of the book."

The work begins with three chapters on the seed and seedling, followed by two upon roots, three upon stems, others upon form and arrangement of buds and leaves and upon the minute structure and the function of leaves. The remaining chapters of Part I deal with the flower and fruit. In Part III, in addition to the rewritten chapters on topics usually found under the caption of ecology, there is a chapter on "Plant breeding" and one on "Some useful plants and plant products." These are the best statements of these topics that have appeared in elementary texts of botany, and constitute a valuable addition to the work as previously outlined in *Foundations of botany*.

Part II is a detailed statement of existing knowledge of the evolution of plants, insofar as that knowledge is based upon morphology and cytology. The second paragraph in this part reads as follows:

One department of morphology (comparative morphology) deals with the various forms or disguises which the same sort of organ may take in different kinds of plants,

<sup>1</sup> BERGEN, JOSEPH Y., and DAVIS, BRADLEY M., *Principles of botany*. pp. ix+555. Boston: Ginn and Company. 1906.

and compares these structures with one another. For example, the foliage leaf is a well-defined organ which can be recognized at a glance; but it requires some study to understand that the scales on the bud and around an onion, and also some forms of spines and tendrils are morphologically leaves, that is, are leaves variously modified. Because all of these structures are related to one another, they are called *homologous*, and morphology studies the *homologies* or relationships of organs. Comparative morphology is one of the interesting subjects of biological study, since it furnishes the basis for the established belief in the evolution or development of the higher plants and animals from simpler forms.

Furthermore, before beginning the study of types, the author says concerning the process of evolution:

The forms and groups split up into divergent lines which constantly gave off, and are still giving off, new shoots. Thus from a number of trunks in the beginning there have been derived a multitude of smaller branches, and from these in turn have arisen countless twigs. It is impossible to construct accurately these genealogical trees, because the species now living occupy the position of buds on the structure, some relatively low down and some at the highest points, but all at the ends of their respective lines of development.

If the student has come to the second part after a careful study of the first, the foregoing paragraphs possibly may be significant to him. If, as a suggested possibility in the preface, the student should begin his study with the second part, such statements must prove bewildering. Presumably he knows little or nothing of the terms used nor of the structures or organs of which they speak, and it would seem that attempts at this point to show the significance of "comparative morphology" would be lost. Also, within the first dozen pages the difficult topic of photosynthesis, with the outlines of the chemical formulae involved, is presented within the limits of a page. Upon a clear understanding of this topic the significance of much of the subsequent work must depend, and if the chemistry of the process is presented at all, it should be in such a form as to be understood by immature minds.

The number of types used and the detail with which their part in the evolutionary story is presented are far greater than has yet appeared in a text designed for elementary use. Indeed few texts used by college classes are so inclusive. It is difficult to understand just how elementary students are going to see the significance of such discussions as (1) that of reduction of chromosomes in Coleochaete as related to claims of a sporophytic generation in that plant; (2) of the "Imperfect fungi;" (3) of the "Evolution of the flower;" (4) of frequent reference in brackets or otherwise to highly specialized genera that are unexplained in the text. The author suggests that such topics be omitted if thereby the teacher's ideas are met in a better way; but in many cases subsequent discussion involves a knowledge of these features. A summary at the end of each group serves to simplify many of the more difficult points.

A highly commendable feature is the introduction of such topics as "Public health," and "Fossil plants and coal." Frequent suggestions as to economic aspects of plants under discussion add much to the value of this part of the book,

as is also true of Parts I and III. The entire book is profusely and well illustrated, some most helpful diagrams being included. While the book as a whole is too heavy for the average high-school work, it will be almost indispensable as a reference work because of its large amount of information, its abundant illustrations, and its helpful suggestions as to the significance of structures and their relationship to one another. A glossary of terms used and a laboratory guide to accompany the text are in process of preparation by the authors. It is unfortunate that the glossary of terms is not bound with the text including the terms to be explained.

The series of texts prepared by COULTER has also been increased.<sup>2</sup> In former textbooks by Professor COULTER, separate treatment was given to morphology and ecology, with work in physiology less distinctly outlined. Those books, while possibly not so extensively used as some others, certainly served as the strongest stimulus in bringing about the introduction of modern botany to supplant the rather mechanical and less representative work that had prevailed in high schools. The testimony of some teachers indicates that at least for some schools *Plant Structures* represents a course too difficult for the students, and that *Plant Relations* is not sufficiently representative of plant processes. *Plant Studies* is made up essentially of parts of the two preceding books bound together. In the new text the author has attempted to articulate more closely morphology, physiology, and ecology, presenting morphological features more prominently than others.

The first five chapters include a presentation of "the structure, function, and relationship of the most obvious plant organs." The plants selected as types in these chapters are all seed plants, those most familiar to high-school pupils, and most readily examined by inexperienced observers. Following this general discussion, eight chapters are given to a consideration of the great groups of plants, beginning with Cyanophyceae and ending with angiosperms, giving an outline of the typical structures, functions, and habits found in each group. Chapter XIV treats of "Pollination," XV of Seed dispersal," XVI to XVIII of the leading families of monocotyledons and dicotyledons, XIX of "Plant breeding," XX of "Forestry," and XXI to XXIV of "Plant associations."

There are several important points in which this book is conspicuously unlike preceding ones by the same author. Botanical terminology is simplified in form and less frequently used. The types selected and the style in which they are presented results in a much simpler treatment of the plant kingdom than in *Plant Structures*. Frequent interpolation of statements concerning economic uses of plants under consideration adds interest and gives a measure of satisfaction to the student's desire for a knowledge of utility. The far too brief chapters on "Plant breeding" and "Forestry" give but a meager suggestion of these prac-

<sup>2</sup> COULTER, JOHN M., A text-book of botany. pp. ix+365. New York: D. Appleton and Company. 1906.

tical phases of botany. The reduction of the space given to plant associations to thirty pages leaves this topic more nearly within the bounds in which it should be found in a general elementary course in botany. But the most important of all the new features of the book is its general plan of organization: first a general introduction to plant structures and functions by the use of the most accessible and best known plants; second upon this foundation follows a presentation of groups in logical order, a knowledge of structure being that around which a knowledge of use and adjustment is arranged, this being done, however, without classifying the different phases definitely into morphology, physiology, and systematic botany; and thirdly, a presentation of special phases of plant life follows the laying of general foundations.—OTIS W. CALDWELL.

#### Water plants

The literature pertaining to water plants promises to be greatly enriched through a series of studies by GLÜCK.<sup>3</sup> The first volume (unhappily called *Erster Teil*, though a book in itself, the second "part" being likewise an independently paged volume) deals with European Alismaceae, of which 8 species, representing 5 genera, were studied. This study differs from that of SCHENCK, SAUVAGEAU, and others, in that while they have specially treated the anatomical or geographical aspects, the emphasis here is chiefly on the biological side and by experimental methods. This shifting from the static to the dynamic is in harmony with the present trend of investigation. The book will be of interest to the ecologist and morphologist, and has as well a message for the systematist.

Part I of this first volume is descriptive of the experiments. Plants were studied in various relations to water, the cultures approximating all ordinary conditions of the uncultivated state. Typical of the author's methods is his treatment of *Alisma Plantago*. He studied first the land forms and those growing in water of various depths. Records were kept of macroscopic observations, measurements of parts, and peculiarities of behavior. Land-grown seedlings were then subjected to various aquatic conditions, even to submergence at depths as great as four meters. Older land plants were also subjected to similar experiments. Both, under certain conditions, determined by size of plant and amount of stored food, took on the form of leaf usually characteristic of the habitat. In some experiments water plants were transformed to land forms. There were also observations on the influence of habitat on the formation of flowers and fruit, and the conditions in which plants pass the winter.

While the results demonstrate the remarkable plasticity of these forms, there seem, however, to be rather definite limits to their variability. For example, the experiments seem to have established the correctness of the old division of *A.*

<sup>3</sup> GLÜCK, HUGO, Biologische und morphologische Untersuchungen über Wasser- und Sumpfgewächse. Erster Teil: Die Lebensgeschichte der europäischen Alismaceen. 8vo. pp. xxiv+312. pls. 7. figs. 25. Jena: Gustav Fischer. 1905. M 20.

*Plantago* L. into two distinct species, as it was impossible, even by long continued cultures, to transform one into the other.

Upon five of the forms special experiments to determine the influence of light were conducted. In general, however, the variable factor was the water relation, and this of course exercised some influence on numerous other relations. To change the degree or depth of submergence of a plant may not only alter the water relation, but cause as well differences in pressure, light, temperature, air, etc. It seems unfortunate that in all this study there was no exact attack upon certain of these factors individually, instead of the consideration of *ensemble* effects. Anatomical studies, too, would have been of interest, but the author felt he could not undertake the task at this time.

Part II is a summary of results. The plants are classed, without regard to systematic relations, into four habitat groups, viz.: land forms, shallow water forms, floating forms, and submerged forms. Each of these habitat groups is characterized by a certain type of leaf, though they of course intergrade. Each of these four dominant leaf types is discussed in relation to the environment producing it, especially the water relation. The optimum condition for general development was usually found to be shallow water. One plant, however, *Echinodorus ranunculoides repens*, grew best on land.

In discussing the flowering habits of aquatic plants the author notes that the land and shallow water forms flower more freely, and he suggests a relationship between the leaf types and the flowering habits. It would seem, however, that the explanation might be readily thrown back, at least to the factors that produced the various leaf types.

Three of the Alismaceae studied were able to develop flowers and to open them while wholly submerged. On bright days these flowers were observed to open in the water, each surrounded by a bubble of gas. Only one of these, *A. graminifolium*, was able to develop and ripen seeds without coming to the surface.

Metamorphosis of flowering shoots into leafy axes was accomplished by changing the habitat. In *Elisma natans* and *Echinodorus repens* the amount of metamorphosis was immediately correlated with degree of submergence. Beyond a certain depth only vegetative structures were developed.

Under the head of formative factors the chief external influences discussed are those of water as such, depth of water, air, temperature, and light. The consideration of internal factors is limited to one—the quantity of reserve food.

The author concludes with a summary, in Latin, of his systematic conceptions of the forms studied.—ROBERT B. WYLIE.

The second volume<sup>4</sup> is concerned with the Utricularias of central Europe, the formation of turions by water plants, and Ceratophyllum. GLÜCK finds,

<sup>4</sup> GLÜCK, HUGO, Biologische und morphologische Untersuchungen über Wasser- und Sumpfgewächse. Zweiter Teil: Untersuchungen über die mitteleuropäischen Utricularia-Arten; über die Turionenbildung bei Wasserpflanzen, sowie über Ceratophyllum. 8vo. pp. xviii+256. pls. 6+figs. 28. Jena: Gustav Fischer. 1906. M. 18.

apparently with some surprise, that the morphology of *Utricularia* shoots can be best understood by considering the leaf and the axis homologous and equivalent structures—a point of view perhaps not so novel to others. Various forms, dependent on habitat, are described in the six recognized species. The so-called rhizoids, really modified shoots, arising from the base of the inflorescence, are partly anchoring and partly nutritive organs. Aerial shoots from water forms assist in gaseous exchange.

The formation and behavior of turions (special buds which become detached) is examined in a considerable range of water plants (*Elodea canadensis*, *Stratiotes aloides*, *Hydrilla verticillata*, *Myriophyllum verticillatum*, *Utricularia* (6 spp.), *Aldrovandia vesiculosa*, *Caldesia parnassijolia*, *Potamogeton* (7 spp.), and *Hydrocharis morsus-ranae*), and the structures are figured. The formation of these buds, usually in autumn in consequence of lowered temperature, may also be brought about by other unfavorable conditions. Growth may be resumed in autumn, but usually they hibernate in the water. Some withstand freezing in ice or mud; most are killed by it. As reproductive and hibernating structures the turions replace seeds largely, since the habitat of water plants is unfavorable to seed-forming.

Regeneration phenomena examined in *Utricularia* were referred to "correlation," the accumulation of plastic material at certain points causing the *Neubildung*.

*Ceratophyllum*, which has been widely believed to be a free swimming plant, is shown to be originally anchored by "rhizoids" (peculiarly modified shoots 6–25<sup>cm</sup> long), which are also permeable to food materials.

New forms are described and the general results of the study are applied to the taxonomy of the plants.

Want of an index in both volumes, beyond mere names of plants, makes the facts gathered difficult of access. When will authors learn that they owe to themselves, if not to other users of their work, the drudgery of index making?—C. R. B.

#### MINOR NOTICES

**Contributions of U. S. National Museum.**—The current issue of this series contains the fifth paper by ROSE<sup>5</sup> under the title "Studies of Mexican and Central American plants." It also represents the fifth journey of Dr. ROSE to Mexico, which has enabled him to write with a large field experience. The numerous plates, some of them reproductions of photographs, bring the plants vividly to the eye. Four new genera are described, as follows: *Calibanus* (Liliaceae), *Sphinctospermum* (Viciaceae), *Pseudoxalis* (Oxalidaceae), *Escontria* (Cactaceae). The four following genera have been reestablished: *Beaucarnia* (Liliaceae), *Odonia* (Viciaceae), *Biophytum* (Oxalidaceae), *Terebinthus* (Balsameaceae).

<sup>5</sup> ROSE, J. N., Studies of Mexican and Central American plants. No. 5. Contrib. U. S. Nat. Herb. 10: 79–132. pls. 16–43. 1906.

The Mexican species of the following genera have been brought together: Beaucarnea (3 new), Dasyllirion (1 new), Nolina (3 new), Castalia (1 new), Benthamantha (3 new), Dolicholus (3 new), Odontia (3 new), sessile-flowered species of Parosela (5 new), some Mexican species of Ionoxalis (17 new), Lotoxalis (3 new), Terebinthus (7 new). The North American species of Krameria are presented, 3 being new. New species are also described under the following additional genera: Echeandia, Clematis (2), Potentilla, Alchemilla (2), Pithecellobium, Bauhinia (3), Cassia (4), Hoffmannseggia, Cologania (2), Geranium (3), Linum (2), Polygala (3), Abutilon, Wissadula (2), Hypericum (4), Calceolaria, Opuntia, Echinocactus, Arracacia (2), Deanea (4), Eryngium (2), Prionosciadium.—  
J. M. C.

#### NOTES FOR STUDENTS

**Plant diseases.**—JONES and MORSE have published further results of their studies of the Irish-potato diseases in Vermont.<sup>6</sup> Spraying late potatoes with Bordeaux mixture for late blight and rot, due to *Phytophthora infestans*, gave an average gain per acre, during fifteen years and covering a number of varieties, of 119 bushels. Against early blight, due to *Alternaria solani*, three sprayings with Bordeaux gave very much increased yields. When loss from rot is anticipated, potatoes should be exposed as long as possible to the sun and air before being placed in storage. Cold storage was found to reduce greatly the damage from rot during storage. Liming and formalin disinfection were of no value. Against potato scab, due to *Oospora scabies*, formalin gas proved effective, even better results secured in this manner than by soaking in formalin solutions.

STEWART, EUSTACE, and SIRRINE have published the results of the fourth year of their ten-year spraying experiment with potatoes.<sup>7</sup> Soda-Bordeaux did not prove more satisfactory than the usual lime-Bordeaux. In thirteen of the experiments reported the average gain due to spraying was \$20.04 per acre, while the average cost of spraying for the same experiments was \$4.25 per acre.

COBB presents further studies on the gumming of sugar cane,<sup>8</sup> a bacterial disease, due to *Bacterium vascularium* (Cobb) Greig-Smith, first described by the same author from Australia. One of the prominent symptoms of the disease is the escape of a yellowish slime or gum from the cut end of the stem. The disease affects chiefly the vascular bundles, but the parenchymatous tissues are also attacked, and in the upper part of the stem cavities, which contain as

<sup>6</sup> JONES J. R. and MORSE, W. J., Potato diseases and their remedies. Rept. Vt. Exp. Stat. 18:272-291. 1906.

<sup>7</sup> STEWART, F. C., EUSTACE, H. J., and SIRRINE, F. A., Potato spraying experiments in 1905. Bull. N. Y. Geneva Exp. Stat. 279:151-229. pl. I-5. map I. 1906. See also Bull. N. Y. Geneva Exp. Stat. 264:93-204. pl. I-16. map I. Rev. in Bot. GAZETTE 41:364. 1906.

<sup>8</sup> COBB, N. A., Third report on gumming of the sugar-cane. Bull. Div. Path. and Phys. Hawaiian Sugar Planters' Assoc. 3:1-46. figs. I-12. 1905.

much as a teaspoonful, are often filled with this yellow slime. Some sorts of cane seem to be quite resistant to the disease, and in their use, as well as in taking care to avoid diseased cuttings or "seed cane," are to be found the principal means of combating the disease.—E. MEAD WILCOX.

Items of taxonomic interest.—J. HUBER (Boletim Mus. Goeldi 4:510-619. 1906), in his sixth paper on the plants of the Amazons, describes a new genus (*Browncepsis*) of Leguminosae (Caesalpinae).—F. S. COLLINS (Rhodora 8:189-196. 1906), in presenting a synopsis of the species of *Acrochaetium* and *Chantrenia* in N. Am., describes 2 new species of the former.—B. L. ROBINSON (*idem* 196-199), in discussing the nomenclature of the New England Lauraceae, shows that under the Vienna rules "spice bush" bears the name *Benzoin aestivale* (L.) Nees, and "sassafras" the name *Sassafras variifolium* (Salisb.) Ktze., thus happily getting rid of two "duplicate monomials;" and in the same issue (202-204) he shows that the well-known "queen of the prairie" (*Spiraea lobata*) becomes *Filipendula rubra* (Hill) Robinson.—J. C. ARTHUR (Bull. Torr. Bot. Club 33:513-522. 1906) has described new species of Uredineae under *Uromyces* (2), *Puccinia* (2), *Melampsora*, *Uredo* (3), *Caeoma*, and *Aecidium* (3).—W. H. BLANCHARD (Rhodora 8: 169-180. 1906) has published 9 new species of *Rubus* from Maine.—M. L. FERNALD (*idem* 181-185) has published a new species and several new varieties of *Carex* from eastern N. Am.—D. PRAIN (Annals of Botany 20:323-370. pls. 24-25. 1906) in a revision of *Meconopsis* recognizes 27 species, four being described as new; and in a revision of *Cathartia* he recognizes 4 species, one of which is new.—In a fascicle of 27 papers on Weberbauer's collections of Andean plants, edited by IGN. UNBAN (Engler's Bot. Jahrb. 37:503-646. 1906), a large number of new species are described, and new genera are established in Leguminosae (*Weberbauerella*) by E. ULRICH and in Asclepiadaceae (*Steleostemma*, *Schistonema*, *Pentacyphus*, *Tetraphysa*, *Stelmatocodon*) by R. SCHLECHTER.—P. DIETEL (Ann. Mycol. 4:421-423. 1906) has described a new genus (*Chnoopsonia*) of Uredineae from India.—T. D. A. COCKERELL (Nature 75: 7. 1906), in a note on "the evolution of the Colorado spiderwort," incidentally describes and names a new species (*Tradescantia universitatis*).—J. M. C.

Scottish peat mosses.—Under a grant from the Royal Society of Edinburgh, FRANCIS J. LEWIS has been investigating the plant remains in the Scottish peat mosses, and some of the results are now published.<sup>9</sup> In the southern uplands the peat in all the districts examined shows a definite stratification of plant remains, indicating a swing from woodland to heath and moss, and again to woodland. In some districts an arctic plant bed is interposed between the lower and upper woodland beds. The regularity of the sequence of the beds

<sup>9</sup> FRANCIS, J. LEWIS, The plant remains in the Scottish peat mosses. I. The Scottish southern uplands. Trans. Roy. Soc. Edinburgh 41:699-723. pls. 6. 1905. II. The Scottish Highlands. *Idem* 45:335-360. pls. 4. 1906.

The history of the Scottish peat mosses and their relation to the Glacial period. Scottish Geog. Mag. 1906:241-252.

and their general agreement on similar although widely separated areas tend to show that they represent successive changes in the vegetation due to climatic changes during the passing away of the Glacial period. None of the districts investigated show any remains of arctic plants at the base of the peat; but some of them, on the contrary, contain remains of hazel in the basal layer. As an illustration of sequence, that of the Galloway peat is as follows: (1) birch forest (lower buried forest), (2) arctic plant beds, (3) pine forest (upper buried forest). The evidence seems to give support to the view that the later phases of the Glacial epoch were separated by somewhat long genial interglacial periods. The details given in the two papers, and well illustrated, are full of interest to students of the succession of floras.

In the Highlands the peat deposits are divided into two groups: (1) those of the western districts, possessing no arctic plants at the base, but a forest bed (*Betula* and *Corylus*), overlaid by plants indicating wet moorland conditions; and (2) those of the northern and northeastern districts, possessing only one well-marked forest bed, and with an arctic plant bed at the base of the peat.—  
J. M. C.

**Geophilous species of Peperomia.**—HILL<sup>10</sup> has been investigating the peculiar geophilous species of *Peperomia*, his attention having been called to them during travels in the Andes of Bolivia and Peru. These species form a comparatively small section of a large genus, having corms or tubers or rhizomes, which seems to be correlated with the possession of more or less peltate leaves. Detailed descriptions of the seedlings of the different species serve as an introduction to general discussion and conclusions. In these forms the cotyledons exhibit a remarkable division of labor, one remaining within the seed and serving as an absorbing organ, the other becoming aerial and green. From this fact as a start, HILL sees change of function of one of the cotyledons gradually leading into the monocotyledonous condition. The large conclusion is the origin of the monocotyledons from dicotyledons, as held by Miss ETHEL SARGANT, but the method is different. Miss SARGANT sees the origin in a phylogenetic fusion of the two cotyledons; while HILL sees it in the change of function of one of the aerial cotyledons to an absorbing organ. While Miss SARGANT traces the connections between *Ranunculaceae* and *Liliaceae*, HILL traces them between *Piperaceae* and *Araceae*. The *Piperaceae* are certainly very primitive dicotyledons, and the *Araceae* are the most Dicotyledon-like of the monocotyledons; and the transition from the geophilous species of *Peperomia* to those of *Arisaema* seems quite plausible. However, such discussions as yet are only suggestive of further investigations; and it is conceivable that there may have been more than one way of passing from the dicotyledonous to the monocotyledonous condition.—J. M. C.

<sup>10</sup> HILL, ARTHUR W., The morphology and seedling structure of *Peperomia*, together with some views on the origin of Monocotyledons. *Annals of Botany* 20: 395-427. *pls. 29-30.* 1906.

**Water movement and turgor in wilting plants.**—PRINGSHEIM has undertaken some important work which may be reasonably expected finally to bring us nearer an understanding of the movement of water in plants. The paper is quite long as compared with the experimental data, though this is perhaps justified by the pioneer nature of the investigation.<sup>11</sup> It seems to be quite universal in cases of wilting that the younger parts are favored in water supply at the expense of the older organs. Investigation proves that there is an actual passage of water from the basal to the apical portions, and that the latter transpire at a relatively greater rate. This movement is associated with a difference in osmotic potential between the base and the apex. The more rapid transpiration of the apical parts increases the concentration of the sap in that region, which is relieved by a movement of water to that locality. This gradient of sap-density from base to apex is correlated with a corresponding turgor gradient. Such facts lead the author to take issue with WIESNER, who has regarded a movement of water as the determining factor in the emergence of adventitious organs. In such cases the author finds an increase in the concentration of the sap in those tissues from which the new parts emerge, which of course necessitates a movement of water in that direction. The difference of osmotic potential in a wilting shoot is, qualitatively, initially present and in no case did it appear where not initially present. While the law of mass action undoubtedly operates to regulate the turgor, the author regards this an inadequate explanation for the relatively high constancy of turgor found.—RAYMOND H. POND.

**Growth and respiration during winter.**—SIMON has studied what he calls the separate growth functions (shooting of buds, elongation of roots, callus-formation, secondary thickening, etc.) and respiration during the period of winter rest.<sup>12</sup> In some cases the suspended activity is autogenous, that is, not necessitated by external conditions. This is true of buds which rest only one season. Those which rest for several seasons do so because the external conditions are not suitable for their growth. If at any time during their long rest the proper conditions are supplied, growth activity is at once resumed. Such a period of inactivity is therefore called aitogenous. The rest of a majority of the meristematic tissues is autogenous. The reduced elongation of the root is partially autogenous, but to a greater extent aitogenous, while callus-formation is entirely the latter. Respiration does not show a period of rest corresponding to that of the growth functions. Under favorable conditions it may be relatively intense. If the temperature be raised to about 22° C. the intensity of respiration is found at any time during the rest period to be only about 25 per cent. less than at the time of its maximum, which is during the active season of the cambium. Res-

<sup>11</sup> PRINGSHEIM, ERNEST, Wasserbewegung und Turgorregulation in welkenden Pflanzen. *Jahrb. Wiss. Bot.* 43:89-144. 1906.

<sup>12</sup> SIMON, SIEGFRIED, Untersuchungen über das Verhalten einiger Wachstumsfunktionen sowie der Atmungstätigkeit der Laubhölzer während der Ruheperiode. *Jahrb. Wiss. Bot.* 43:1-48. 1906.

piration reaches its minimum just before the cambium resumes its activity. The paper includes a rather extended inspection of the literature.—RAYMOND H. POND.

**Ecology of woodland plants.**—WOODHEAD<sup>13</sup> has investigated the woodland plants near Huddersfield, England, and has made an important addition to the ecological literature that is rapidly developing in Great Britain. The subject is still new enough, however, for the author to preface his paper by a brief account of the study of ecology in Britain. The first section of the paper is physiographic, dealing with the woodland vegetation maps of the various areas under consideration. The second section is anatomical, discussing the effect of environment on structure. Under the head of dominant species, there are described the leaf, petiole, and rhizome of *Pteris aquilina*; the leaf, absciss-layer, and elongated bulbs of *Scilla festalis*; and the leaves of *Deschampsia flexuosa*, *Holcus mollis*, and *Vaccinium Myrtillus*. Under the head of secondary species, the leaves of *Heracleum Sphondylium*, *Lamium Galeobdolon*, and *Mercurialis perennis* are described. The vegetation of the Huddersfield district is naturally divided into three parallel zones: (1) the moss moor (1700–1000 feet altitude), the most exposed zone, dominated by *Eriophorum vaginatum* or on drier ridges by *Vaccinium Myrtillus*; (2) the millstone-grit plateau (1000–500 feet), with oak as the dominant tree, associated with birch and pine; (3) the Coal-measure area (500–200 feet), with deep and well-watered humus soil.—J. M. C.

**Phycophaein.**—TSWETT finds<sup>14</sup> that phycophaein does not exist as such in the living body of the brown algae. He does find, however, that a colorless chromogen, soluble in water but insoluble in alcohol, is present. An extract made with distilled water contains this chromogen, but owing to its neutral reaction the solution becomes colored very slowly. Extraction with an alkaline solvent, such as ordinary tap water, gives a yellow solution, rapidly becoming brown by oxidation. Decoloration occurs when the reaction is made acid. Extracts of the thallus with 50 to 80 per cent. alcohol remain colorless. This work, done in REINKE's laboratory, sustains his hypothesis of the *post-mortem* origin of phycophaein and refutes the generally accepted notion that it is a genuine pigment of the chromatophore. The remainder of the paper outlines the author's method for isolating the several pigments present in the chromatophore. The following are soluble in petroleum ether containing 10 per cent. alcohol; carotin, chlorophyllin *a*, fucoxanthin, and fucoxanthophyl. Chlorophyllin  $\gamma$  is not soluble in this reagent, but is soluble in alcohol and ether. The natural color of the brown algae results from a mixture of these pigments in the chromatophore.—RAYMOND H. POND.

<sup>13</sup> WOODHEAD, T. W., Ecology of woodland plants in the neighborhood of Huddersfield. Jour. Linn. Soc. Bot. 37:333–406. figs. 70. 1906.

<sup>14</sup> TSWETT, M., Zur Kenntnis der Phaeophyceenfarbstoffe. Ber. Deutsch. Bot. Gesells. 24:235–244. 1906.

"Droppers" of *Tulipa* and *Erythronium*.—Miss ROBERTSON<sup>15</sup> has been investigating the peculiar descending stolons of *Tulipa* and *Erythronium*, which are called "droppers" by the gardeners. In her account she makes no mention of the work of RIMBACH,<sup>16</sup> one of whose papers contains quite an extended account of the "droppers" of *Erythronium*, and the other deals with a number of genera of the Liliaceae. The summary states that "the power of lateral migration to prevent overcrowding, and of descent into the soil for protection against frost, drought, and animals, is possessed in some degree by many bulbous plants." The statement includes a fact and its purpose; the former seems evident, the latter would be somewhat difficult to demonstrate. The power referred to is said to be more highly specialized in *Tulipa* and *Erythronium* than usual, and hence the structure and behavior of the so-called "droppers" is of special interest "The immature bulb each year produces a single foliage leaf, continued at the base into a hollow tube, the 'dropper,' enclosing a bulb at its tip." It seems to be a distinct device for distributing bulbs; and the testimony of anatomy is that the "dropper" is partly axial and partly foliar.—J. M. C.

Laws of heredity classified.—DARBISHIRE<sup>17</sup> draws a careful distinction between the several so-called laws of inheritance now under discussion among biologists. He classifies them as statistical and physiological according as they are descriptive and deal with mass-results on the one hand, or as they are explanatory and deal with individuals on the other. He places GALTON'S and PEARSON'S laws in the former category, and Mendelism and the law of diminishing contribution in the latter. By the law of diminishing contribution he designates the view generally held that an individual inherits more or less from all his ancestors, but less from the more remote than from the more recent. It is maintained that Mendelism is not to be considered alone as a theory, but also as a method, and that new details of theory which are worked out by the Mendelian method are as much a part of Mendelism as is the original statement made by MENDEL. An experiment is outlined which is calculated to test simultaneously the truth both of GALTON'S and of MENDEL'S laws, and it is pointed out that the former is true of masses and the latter of individuals.—GEO. H. SHULL.

Radial growth of tree-trunks.—LIGNIER<sup>18</sup> has been investigating the growth of trees in diameter, following a suggestion made A. DE CANDOLLE in his *Physiologie végétale* (1832) that if there could be made a very large number of observations of individuals of the same species, an approximate formula of increase

<sup>15</sup> ROBERTSON, AGNES, The "droppers" of *Tulipa* and *Erythronium*. *Annals of Botany* 20: 429-440, pls. 31-32. 1906.

<sup>16</sup> BOT. GAZETTE 30:171-188. pl. 13. 1900; 33:401-420. pl. 14. 1906.

<sup>17</sup> DARBISHIRE, A. D., On the difference between physiological and statistical laws of heredity. *Mem. and Proc. Manchester Lit. Phil. Soc.* 50 (no. 11): 44. 1906.

<sup>18</sup> LIGNIER, O., Notes sur l'accroissement radial des troncs. *Bull. Soc. Linn. Normandie* V. 9:181-224. 1905.

might be established for each species, and it would be possible to estimate with some degree of accuracy the age of a tree from its diameter. From this stand-point LIGNIER has investigated *Quercus pedunculata*, *Castanea vulgaris*, *Sophora japonica*, and *Taxus baccata*; and in a less complete way *Sequoia gigantea*, *Cedrus Deodora*, and *Araucaria imbricata*. There are three distinct periods in the rate of diameter increase: (1) the period of acceleration; (2) the period of decline; (3) the final period in which diameter increase is barely perceptible. *Quercus* begins its final period at about 140 years, when the radius is 57.9 cm.; *Castanea* at 190 years, with a radius of 74.7 cm.; *Taxus* at 150 years, with a radius of 25.6 cm. For each of the species investigated there is a table which gives the age in terms of the radius.—J. M. C.

**Cytology of the Entomophthoraceae.**—The preliminary announcement of RIDDELL'S results was noted in this journal.<sup>19</sup> The full paper has now appeared,<sup>20</sup> *Empusa Grylli* and several species of Entomophthora were investigated, and the writer reaches conclusions somewhat different from those of OLIVE.<sup>21</sup> The nucleus is well developed, there being a chromatin nucleolus surrounded by chromatin granules. At division there is a well-developed mitosis, in which, however, the chromosomes are formed by a direct aggregation of chromatin granules without the formation of a spirem. The spindle is intranuclear, bipolar, and without centrosomes. The conidia of *Empusa* are multinucleate and those of Entomophthora uninucleate. The zygospores of Entomophthora are formed by the fusion of multinucleate hyphal bodies. In *Empusa* the zygospores are formed by the budding out of a hyphal body. Cytological evidence favors the derivation of the Entomophthoraceae from a Mucor-like ancestry.—CHARLES J. CHAMBERLAIN.

**Parthenogenesis in Wikstroemia.**—WINKLER'S preliminary paper was reviewed in this journal,<sup>22</sup> and now the completed results have been published.<sup>23</sup> The present investigation shows that the embryo develops from the egg without fertilization, but whether the egg contains the sporophyte number of chromosomes (52) or the gametophyte number (26) was not determined definitely. It is probable that the egg has the sporophyte number of chromosomes. WINKLER proposes the phrase *somatic parthenogenesis* for cases in which the embryo develops from an egg with the sporophyte number of chromosomes, and *generative parthenogenesis* for cases in which the number has been reduced. He insists that an egg is an egg whether it has the reduced number of chromosomes or not. The

<sup>19</sup> BOT. GAZETTE 42:236. 1906.

<sup>20</sup> RIDDELL, LINCOLN W., On the cytology of the Entomophthoraceae. Proc. Amer. Acad. 42:177-197. pls. 1-3. 1906.

<sup>21</sup> BOT. GAZETTE 41:192-205, 229-259. 1906.

<sup>22</sup> BOT. GAZETTE 39:236. 1905.

<sup>23</sup> WINKLER, HANS, Ueber Parthenogenesis bei *Wikstroemia indica*. Ann. Jard. Buitenzorg II. 5:208-276. pls. 20-23. 1906.

paper closes with an interesting discussion of the significance of the reduction of chromosomes and its relation to alternation of generations.—CHARLES J. CHAMBERLAIN.

**Legume inoculation.**—KELLERMAN and ROBINSON<sup>24</sup> have been investigating the conditions under which a particular species of legume might be successfully inoculated, and the conditions under which failure to obtain inoculation might be expected. A summary of the results is as follows: lime is of decided benefit in obtaining successful inoculations of legumes in some soils (often showing an acid reaction to litmus); soil extracts serving as culture media often indicate the probable success of inoculating a leguminous crop; at least during the first season's growth no general cross-inoculation takes place (bacteria from one host, however, may inoculate a physiologically related species); heavy inoculation by a pure culture increases nodule formation if the soil solution is enriched by the excess of culture medium (in a favorable soil, however, a light inoculation well-distributed is as effective); thorough aeration is favorable to nodule formation.—J. M. C.

**Rate of growth of "fairy rings."**—In view of the fact that little is known of the length of the life cycle of most of the Basidiomycetes, an observation of THOMAS<sup>25</sup> on *Hydnellum suaveolens* is of interest. This fungus forms "fairy rings" in forests. Such a ring was observed by THOMAS in 1896, and at that time the radius of the circle was 8.41 m. The sporophores of the fungus appeared irregularly during the following years, but at no time did those of *H. suaveolens* appear within the circular area once occupied by the mycelium of that fungus. In the years 1901, 1902, and 1905 it was again possible to make radial measurements, when the radius of the circle was 9.54 m., 9.92 m., and 10.56 m., respectively. From the annual increase thus obtained the calculated age of the circle was about 45 years. For nearly half a century the mycelium had grown, spreading over the area of a fairy ring scarcely 20 m in diameter.—H. HASSELBRING.

**Seedlings of gymnosperms.**—HILL and FRAINE have been studying the anatomy of the seedlings of Coniferales, and an abstract of their paper has been published.<sup>26</sup> The general conclusions are that the cotyledonary bundles of *Cephalotaxus* and *Taxus* exhibit mesarch structure; that the gymnosperms as a whole exhibit three varieties of rotation, namely (1) that in which the cotyledonary bundle is endarch throughout and the rotation of the protoxylem is very indefinite (as *Cephalotaxus*), (2) that in which the cotyledon-trace is endarch but the rotation of the protoxylem takes place in the hypocotyl (as *Cedrus*),

<sup>24</sup> KELLERMAN, KARL F., and ROBINSON, T. R., Conditions affecting legume inoculation. U. S. Dept. Agric., Bur. Pl. Ind., Bull. 100, part viii. pp. 15. pls. 2. 1906.

<sup>25</sup> THOMAS, FR., Die Wachstums geschwindigkeit eines Pilzkreises von *Hydnellum suaveolens* Scop. Ber. Deutsch. Bot. Gesells. 23:476-478. 1906.

<sup>26</sup> HILL, T. G., and FRAINE, E. DE, On the seedling structure of gymnosperms. Annals of Botany 20:471-473. 1906.

and (3) that in which the rotation of the xylem and bifurcation of the phloem of the cotyledonary bundle take place in the cotyledon (as *Pinus*); and that the numerous cotyledons obtaining in many plants have been formed by the splitting of the preexisting ones.—J. M. C.

**Plant formations at Victoria Falls.**—Miss GIBBS<sup>27</sup> has published an enumeration of the plants collected in two localities in southern Rhodesia during a period of three months. The list is a long one, and the new species are numerous. A second part of the paper describes the plant formations in the vicinity of Victoria Falls, three distinct regions being recognized: (1) The veldt, or open forest growth, which is found throughout southern Rhodesia, extending on both sides of the Zambezi River as far as the eye can see; (2) a region limited to the immediate banks of the Zambezi and the islands above Victoria Falls, in which *Eugenia guineensis* is dominant; (3) a region including the bog edge of Livingstone Island and that of the rainy forest in general, dominated by *Eugenia cordata*. The four plates reproduce eight photographs of characteristic plants and plant formations.—J. M. C.

**Aluminum in soil and water cultures.**—ROTHERT<sup>28</sup> gives in preliminary form the work done by his pupils on the general relation of plants to aluminum.<sup>28</sup> The soluble salts of this metal and also to some extent the insoluble phosphates will enter the plant from soil and water cultures. Entrance to the plant occurs much more readily from the water than from the soil cultures. After entering the roots the solutes do not migrate to other tissues but accumulate in the tissues of the root, so that on analysis nearly all and in some cases all of the metal found is in the roots. Very dilute solutions were found to stimulate growth, while the more concentrated ones were toxic.—RAYMOND H. POND.

**Conifers of China.**—MASTERS<sup>29</sup> has enumerated the conifers of China, which is of special interest since the China-Japan region contains more endemic genera of conifers than any other. The 89 species recognized are distributed among 21 genera as follows: *Podocarpus* (7), *Dacrydium*, *Cephalotaxus* (6), *Torreya* (2), *Taxus*, *Pinus* (14, with 2 new species), *Larix* (7), *Pseudolarix*, *Picea* (15, with 3 new species), *Tsuga* (5), *Pseudotsuga*, *Keteleeria* (4), *Abies* (8, with a new species), *Cunninghamia*, *Taiwania*, *Cryptomeria*, *Glyptostrobus*, *Libocedrus*, *Thuja* (2), *Cupressus* (3), *Juniperus* (7).—J. M. C.

**Freezing.**—What kills a plant when it freezes? Various answers have been made to this question, the current one (PFEFFER, JOST) being that death is due

<sup>27</sup> GIBBS, Miss L. S., A contribution to the botany of southern Rhodesia. *Jour. Linn. Soc. Bot.* 37: 425-494. *pls. 17-20.* 1906.

<sup>28</sup> ROTHERT, W., Das Verhalten der Pflanzen gegenüber dem Aluminium. (*Vor. Ber.*) *Bot. Zeit.* 64: 43-52. 1906.

<sup>29</sup> MASTERS, MAXWELL T., On the conifers of China. *Jour. Linn. Soc. Bot.* 37: 410-424. 1906.

to the withdrawal of water. GORKE goes a step further and shows<sup>30</sup> that the low temperatures besides producing various analogous changes, lead to intramolecular transpositions in the proteids, which are generally accompanied by a precipitation of a part of the dissolved ones; and when such chemical alternations surpass a definite limit death ensues.—C. R. B.

**Sap-flow.**—WIEGAND gives a useful résumé of the facts and theories in regard to the flow of the sap from bleeding trees, especially the maple, to which he adds some observations of his own. He holds that the bleeding due to pressure of expanding gases is inadequate to account for the volume of sap exuded, and that the best explanation is the one that ascribes the pressure to the excretion of water by cells of the medullary rays, stimulated to activity by rising temperature.—C. R. B.

**Glycogen and paraglycogen.**—Since MASSART's edition of ERRERA's posthumous paper on this subject,<sup>32</sup> drawings have been found and copied with "scrupulous fidelity." These are now issued as a supplementary paper.<sup>33</sup> The five plates are handsome chromolithographs showing the distribution of glycogen and paraglycogen in the rhizopods and flagellates, as well as several groups of fungi, including the Myxomycetes.—RAYMOND H. POND.

**Cotyledons of *Trollius*.**—RAMALEY<sup>34</sup> has found that the petioles of the cotyledons of *Trollius albiflorus* are united for about half their length, the plumule escaping through a slit in this sheath. LUBBOCK described a similar condition in *T. Ledebourii*, and it seems to be general throughout Ranunculaceae.—J. M. C.

**Mycorrhiza and nitrogen fixation.**—Contrary to the supposition of MÜLLER, MÖLLER finds that the dichotomous mycorrhiza of the mountain pine is of no use in fixing free nitrogen for the tree.<sup>35</sup>—RAYMOND H. POND.

**Anatomy of roots.**—HOLM<sup>36</sup> has described and illustrated the anatomical structure of the roots of species of *Spigelia*, *Phlox*, and *Ruellia*.—J. M. C.

<sup>30</sup> GORKE, H., Ueber chemische Vorgänge beim Erfrieren der Pflanzen. Landw. Versuchs-Stat. 65:149-160. 1906.

<sup>31</sup> WIEGAND, K. M., Pressure and flow of sap in the maple. Amer. Nat. 40: 409-453. 1906.

<sup>32</sup> See review in BOT. GAZETTE 41:370. 1906.

<sup>33</sup> ERRERA, LEO, Dessins relatifs au glycogen et au paraglycogen. Recueil de l'Institut bot. Bruxelles 1:432-436. pls. 5. 1906.

<sup>34</sup> RAMALEY, FRANCIS, The seed and seedling of the mountain globe-flower. Univ. Colorado Studies 3:93-95. figs. 13. 1906.

<sup>35</sup> MÖLLER, A., Mykorrhizen und Stickstoffnährung. Ber. Deutsch. Bot. Gesells. 24:230-233. 1906.

<sup>36</sup> HOLM, THEO., The root-structure of *Spigelia marilandica* L., *Phlox ovata* L., and *Ruellia ciliosa* Pursh. Amer. Jour. Pharmacy 78:553-559. figs. 5. 1906.

## NEWS

DR. E. GILG, curator of the Botanical Museum, has been appointed to a professorship in the University of Berlin.

PROFESSOR W. A. KELLERMAN, Ohio State University, will soon make a third collecting trip to Guatemala for the purpose of collecting parasitic fungi.

ALEXANDER W. EVANS, Yale University, has been promoted from an assistant professorship to the Eaton professorship of botany in the Sheffield Scientific School.

A NEW WORK on the botany of California is announced by C. R. Orcutt under the title "California Plants." It is to be issued in parts, each containing descriptions of about 100 species.

MEL T. COOK, recently in charge of the Department of Plant Pathology of the Central Agricultural Experiment Station of Cuba, will spend the next few months at the New York Botanical Garden.

DR. ERNEST PFITZER, who has been professor of botany and director of the Botanic Garden in the University of Heidelberg since 1872, died on December 3, at the age of sixty-one. His researches lay in the field of anatomy and physiology.

DR. W. B. MACCALLUM, assistant in plant physiology in the University of Chicago, has been appointed professor of plant physiology in the University of Arizona at Tucson. He entered upon his new duties on January 1, which involve no teaching, but solely research. The University of Arizona in establishing a research position in this line has shown the way to many older institutions.

OWING to the amount of material which has been received for publication, the *Philippine Journal of Science* will be issued in three sections, each separately paged and indexed: (a) General Science, (b) Medical Sciences, (c) Botany. The section on botany will include all botanical work issued from the Bureau. It will not be issued at regular monthly intervals, but as fast as material is available: yet each year's subscription (\$2) will cover a sufficient amount of material to constitute a fair-sized volume.

# BOTANICAL GAZETTE

FEBRUARY 1907

## POLLEN DEVELOPMENT IN HYBRIDS OF *OENOTHERA LATA* $\times$ *O. LAMARCKIANA*, AND ITS RELATION TO MUTATION<sup>1</sup>

## CONTRIBUTIONS FROM THE HULL BOTANICAL LABORATORY LXXXIX

REGINALD RUGGLES GATES  
(WITH PLATES II-IV)

This paper is a record, in part, of some investigations upon DeVRIES' mutants of *Oenothera*. The work was begun at the Marine Biological Laboratory, Woods Hole, Mass., in the summer of 1905; continued in the Botanical Laboratory of McGill University during the year 1905-6; and again at Woods Hole in the summer of 1906, where a larger garden of evening primroses was grown and studied, and material collected for further work; which is now being prosecuted at the Hull Botanical Laboratory, University of Chicago.

I have to thank Dr. B. M. DAVIS, under whose careful guidance the research was begun, for much attention and many suggestions and criticisms. I also have pleasure in acknowledging my indebtedness to Professor D. P. PENHALLOW, McGill University, for placing all the facilities of the Botanical Laboratory at my disposal, and for many kindly suggestions, to which Assistant Professor CARRIE M. DERICK also contributed, as the work progressed. The investigations at Woods Hole were carried on with the aid of scholarships from McGill University. I have to thank Dr. GEORGE T. MOORE for encouragement in the work of last summer, and also

<sup>1</sup> A preliminary paper was presented before Sections F and G of the American Association for Advancement of Science at New York, December 28, 1906.

Mr. GEO. M. GRAY for aid in planting and caring for the garden. These researches are being continued with the aid of a fellowship at the University of Chicago, and I am indebted to Professor JOHN M. COULTER and Dr. CHARLES J. CHAMBERLAIN for supervision.

#### Materials and methods

The first material of *Oenothera lata* was collected from a small garden of evening primroses at Woods Hole in 1905 from guarded seeds of DeVRIES. Stages were preserved for a study of sporogenesis and embryo sac development in *O. lata* and its hybrid forms; but the results recorded here will concern only pollen development, as the work on the megasporangium and embryo sac has not been completed. The study of pollen development was taken up first because an interesting problem was presented by the abortion of the pollen, and because the reduction divisions in the microspore mother cell are for various reasons more easily investigated than those of the megasporangium mother cell. The possibility of a cytological explanation of the segregation of characters in Mendelian hybrids, no one of which has yet been worked out on a chromosome basis, as well as the possibility of an explanation of the phenomena of mutation itself have also been kept in mind. The work has not yet reached a stage where it is possible to say that a cytological explanation for the Mendelian segregation of characters will be found, though this is possible. Some of the other results which will be described later, however, suggest that there may be found in the chromosomes a basis for the explanation of discontinuous variation or mutation, the suggestion being that the appearance of the mutant is preceded by certain morphological changes in the chromatin of the germ cells from which it originates, or, more exactly stated, that the germ cells from which a mutant arises differ in the structure of their idioplasm from the ordinary germ cells of the parent species.

Material has also been collected from *O. Lamarckiana* and several other of the mutants for comparison with *O. lata*.

The garden of 1906 at Woods Hole contained *O. Lamarckiana* and five of the mutants, all grown from guarded seeds of DeVRIES, which were planted in the greenhouse at the University of Chicago, the young seedlings being transplanted at Woods Hole about May 25.

By July 28, *O. rubrinervis* had begun blooming freely, and the other forms, including *O. lata*, began several days later. The earlier blooming of *O. rubrinervis* was very noticeable.

The history of the growth of these plants, which was observed with some care, will not be taken up here. Numerous crosses were made and other flowers self-pollinated for a study of the next generation, all necessary precautions being taken by means of bags, etc., to guard the flowers and to secure pure fertilization.

As stated by DeVRIES (33), the mutant *O. lata* does not mature its pollen and hence must be pollinated from another species, thus producing a hybrid in the second and succeeding generations. The seeds used were from a cross of *O. lata* with the pollen of *O. Lamarckiana*. These are said by DeVRIES (*l. c.* 1:168) to segregate in the next generation, giving about 15-20 per cent. of *O. lata* plants and the remainder *O. Lamarckiana*, the characters of the latter thus being dominant.

My garden of 1906 contained fifteen plants from seeds of *O. lata* pollinated by *O. Lamarckiana*, ten of which conformed more or less completely to the characters of the pollen parent and four to the *lata* type, which is easily distinguished even in the rosette stage. One plant (no. 79), however, differed markedly from either of these forms, and was clearly a "mosaic" hybrid, *i. e.*, in some characters it resembled one parent and in some characters the other parent. The petals, however, were considerably larger than those of either parent species, and the sepals showed streaks of red, suggesting the sepals of *O. rubrinervis* but much paler. This character, however, is common to all the plants of *O. lata*  $\times$  *O. Lamarckiana* having the *Lamarckiana* characters, and is occasionally seen to a less degree in *O. Lamarckiana* itself. The large ovaries and stout hypanthium, the greater pubescence on the young buds, and the broad leaves with their obtuse tips, are all characters of *O. lata*. But the leaves were scarcely at all crinkled (the more or less complete absence of crinkling being a character of *O. Lamarckiana* which distinguishes it easily from *O. lata*), and the general habit of branching and greater luxuriance of growth also correspond with *O. Lamarckiana*. This plant is mentioned as showing that segregation of the parental characters is not always complete in this cross, as this individual was fairly

intermediate in position between the two parents, though no fractionization of any characters was observed.

In killing material various fixing fluids were used, the most satisfactory being 1 per cent. chromacetic, a chromacetic solution made up as follows: 1 per cent. chromic acid, 70°<sup>c</sup>; glacial acetic acid, 0.5°<sup>c</sup>; water, 30°<sup>c</sup>; and the following chromosmacetic formula: 1 per cent. chromic acid, 70°<sup>c</sup>; glacial acetic acid, 0.5°<sup>c</sup>; 2 per cent. osmic acid, 5°<sup>c</sup>; water, 30°<sup>c</sup>.

Sections were cut varying in thickness from 3  $\mu$  to 10  $\mu$ . The stains producing the best results were found to be Haidenhain's iron-alum-haematoxylin with orange G as a plasma stain, and Flemming's triple stain. The former is most satisfactory for the study of karyokinetic figures, particularly the chromosomes, but safranin and gentian violet give equally good results for synapsis stages.

#### Conditions of degeneration of pollen and tapetum in anther development

Unless otherwise stated, this account will refer to conditions observed in *O. lata*. The morphology of the *Lamarckiana* hybrid is very similar, so that the description would apply equally well to the latter, except where the pollen abortion in *O. lata* causes abnormalities.

*Fig. 1* shows a transverse section of a single loculus in the "resting" pollen mother cell stage. The epidermal cells when normally developed have thick, highly cutinized walls at this early stage. Occasionally, when failure of anther development sets in at an earlier stage this cutinization does not take place, the cells remaining thin-walled. There are several "middle layers,"<sup>2</sup> generally about four. These are narrow radially and often show the effects of pressure from the large cells of the tapetum. The latter forms a well-organized layer of cells enclosing the sporogenous tissue. The granular appearance of the tapetal cells, owing to deeply-staining inclusions which fill the cytoplasm, easily distinguishes them from the middle layers. There are usually one to three longitudinal rows of pollen mother cells in a loculus, but the rows are not regular and a cross-section may show as many as five mother cells. In the great majority of cases the

<sup>2</sup> There is some variation in the use of this term. It is here applied to all the layers between the epidermis and the tapetum, because they are undifferentiated and all have the same developmental history.

tapetum is normal at this time, but occasionally some of the tapetal cells or even the mother cells may be found degenerating in this "resting" condition; in fig. 3 both these conditions are present. Certain scattered tapetal cells show by their loss of turgor and their deeply staining contents that they are degenerating. Their nuclei may become flattened, but at first retain their nucleoli and nuclear wall. Later the latter breaks down and the nucleoli lose their outline and become indistinct, until finally the nucleus has disappeared. These changes are frequently accompanied by vacuolation. The cytoplasm stains progressively deeper with safranin and with haematoxylin. This may be due to the distributed condition of the chromatin, which diffuses through the cell after the nuclear membrane disappears, or to the formation of some acid product in the cytoplasm as the result of the mucilaginous degeneration. Similar staining reactions are shown by degenerating mother cells. Fig. 4 shows a stage of this degeneration.

Most of the recent papers on pollen development in sterile hybrids do not describe the condition of the tapetal cells. GREGORY (9) mentions that the somatic cells surrounding the pollen mother cells in a hybrid race of *Lathyrus odoratus* are normal. JUEL (13), in his description of pollen development in the sterile hybrid *Syringa Rothomagensis*, makes no reference to the tapetal cells; nor does CANNON describe the condition of the tapetum in his two papers (4, 5). TISCHLER (31) finds conditions in the tapetal cells of hybrids of *Ribes* very similar to those described in some detail by BEER (3) in *Oenothera biennis* and *O. longiflora*, and which I have observed also in *O. lata* and *O. Lamarckiana*.

The tapetal cells in *O. lata* are uninucleate during the "resting" stage of the pollen mother cells. During the early presynaptic stages in the mother cells the tapetal nuclei divide mitotically, and the binucleate condition persists for some time. This first nuclear division in the tapetal cells may take place almost simultaneously throughout a loculus, so that many mitotic figures in various stages are often found in the same section. Later divisions are more irregular and are probably chiefly by amitosis, as stated by BEER (3) for *O. longiflora* and *O. biennis*, although occasionally two spindles may be found in the same tapetal cell.

My observations on the later stages of tapetal development agree in the main with those of BEER, and so will not be taken up here in detail. In some cases appearances suggest simultaneous division of a nucleus into several smaller nuclei, the latter often forming closely aggregated masses, as figured by BEER. At a still later stage, when the persistent pollen grains have begun to assume their adult shape, fusion of these masses of nuclei into one or two apparently occurs, and the tapetal cells return to a binucleate or even uninucleate condition. Both nuclei and cytoplasm now, however, show a very different appearance from the early uninucleate and binucleate condition, the nuclei being at this time much larger and having extremely little chromatic content. The cytoplasm, which is loosely reticular rather than granular at this time, frequently contains more or less indefinite and irregular chromatic masses. BEER describes the origin of these masses and compares them with the chromidial structures found by MEVES (19) in the tapetal cells of *Nymphaea alba*, and with similar bodies found by GOLDSCHMIDT (8) in the cytoplasm of certain animal tissues. These tapetal cells of the genus *Oenothera* offer excellent material for a further study of amitosis. It is interesting to note that the structure of the tapetal cells in the mutant *O. lata* is apparently identical with that of the well-known members of the genus, such as *O. biennis*. During these later nuclear divisions of the tapetal cells in *O. lata*, radial elongation of the cells may take place, but rarely enough to nearly fill the cavity of the loculus left by the degeneration (in such cases) of the mother cells. Transverse divisions of the tapetal cells are comparatively rare.

In the great majority of cases the tapetal cells begin to break down before disintegration in the mother cells sets in. As already stated, this disintegration may appear as early as the resting stage of the mother cells, where scattered cells begin to lose their turgor and degenerate. In later stages of development a great variety of conditions of disintegration of this tissue may be found. The tapetum may break down irregularly in different parts of a given loculus, or only on one side, or in patches. Different loculi of the same anther and different anthers of the same flower show wide variations in this matter.

POHL (22) states, followed by DEVRIES (*I.c.* 1:292), that in *O.*

*late* pollen development is normal up to the tetrad stage; that the tapetal cells later elongate and multiply, filling the whole loculus; and that the pollen cells in the mean time have broken down and disappeared, only a few small scattered grains remaining. The first statement is erroneous, because degeneration may take place in the pollen mother cells before the reduction divisions (*figs. 3, 4*). POHL's account of the history of the tapetal cells is also contrary to my observations. It is true in general that the tapetal cells continue active during synapsis and the reduction divisions in the pollen mother cells, although breaking down frequently begins at this time, or even earlier, in the "resting" stage, as already mentioned. But in later stages, when the pollen cells have largely disappeared after the reduction divisions, leaving an empty cavity, the normal condition is not as POHL states—ingrowth of the tapetal cells to fill the cavity—but partial or complete disappearance of the tapetum; and in the latter case *subsequent ingrowth* of the middle layer cells behind to fill more or less completely the cavity (*figs. 5, 6*). Stages of this process are found in which the disintegrating tapetal cells form a thin layer lining the locular cavity (which is usually empty under these conditions), the remains of this layer sometimes appearing later as a thin, homogeneous, deeply staining ring (*fig. 5*), which may afterwards completely disappear. The middle layer cells, which now line the cavity, in the meantime become rounded on their inner surfaces, owing to the release of the pressure of the tapetal cells. They then push in to fill the cavity and may even multiply by mitotic division. The cavity of the loculus may thus become completely closed (*fig. 6*).

POHL's error in mistaking these middle layer cells for tapetum was probably due to a hasty and insufficient examination, for in carefully prepared sections the former have a totally different appearance from the tapetal cells, for which they could not possibly be mistaken. While the latter are easily recognizable by their densely granular contents and conspicuous nuclei, as well as by their rectangular shape, the middle layer cells have no granular contents, are uninucleate, with small nuclei having little chromatin, and are rounded and irregular in shape. POHL's error is probably to be attributed to his failure to observe the degeneration and disappearance of the tapetum before the ingrowth of the middle layer cells takes place, as well as to distin-

guish between the cells of the middle layers and the tapetum by their appearance.

This condition, where the tapetum breaks down early and the middle layer cells then grow in and fill up the loculus, is common and may occur in one loculus of an anther when the adjoining loculi show normal functioning tapetum. The ingrowth is frequently irregular, occurring more rapidly on one side of the cavity, thus causing the latter to lie eccentrically in the anther lobe and to assume a narrow and flattened or irregular shape. This ingrowth of middle layer cells does not initiate the disintegration by pressure on the tapetal cells, or in any other way, for it only begins after the tapetum has more or less completely disappeared, which is long after the first appearance of degeneration in the tapetal cells. That the tapetal cells are necessary for the nourishment and growth of the pollen grains is shown by the entire absence of the latter in such cases long before the cavity is obliterated.

Another and perhaps more common condition is for the tapetum to persist through the tetrad divisions and until the persistent pollen grains attain a large size. The loculus meantime enlarges considerably by growth (*fig. 7*), and also by the flattening and breaking down of the middle layers, as would be expected in normal pollen development. The tapetal cells also generally become flattened at this time, and stretched out so as to line the larger cavity created partly by growth and partly by the degeneration of the middle layers. In such loculi pollen grains in various stages of development are frequently though not always found, although when present they almost invariably assume irregular and bizarre shapes, though they may contain nuclei which are normal in appearance. Rarely pollen grains may be seen which have the triangular disk-shaped appearance characteristic of the pollen in this genus; but such grains have never been observed to reach full size, although abnormally shaped grains may do so sometimes.

It is evident that the break-down of the middle layers, as mentioned in the last paragraph, is due to pressure from the functioning tapetum, for the cells of the middle layer become flattened radially and lose their contents, while the tangential walls may persist for some time, forming irregular concentric rings of dark-staining substance. Later

these completely disappear, leaving no trace of the middle layers. The tapetum then lies directly against the epidermis.

The presence of pollen grains in various stages of development when the tapetum persists, and their complete absence when the tapetum disappears early, shows that the failure of the tapetum to develop is at least one factor in the abortion of the pollen. POHL's conclusion that "die Zellvermehrung und relative Persistenz der Tapetumzellen hemmt die Pollenentwicklung" is evidently an error, for, in the great majority of cases at least, where the tapetum persists, it does not show abnormal radial elongation or growth to fill the locular cavity, the tapetal cells in older stages becoming instead very much flattened radially, as already seen. The cases where the tapetum does not persist, but where the cells of the middle layers grow in and obliterate the cavity, have already been described. In short, when the tapetum persists the middle layers break down, as is the usual condition in normal pollen development; but when the tapetum degenerates at an abnormally early stage, the cells of the middle layers grow in and fill the loculus.

#### History of the pollen mother cells

##### PROPHASE AND SYNAPSIS

The description thus far has dealt chiefly with the general morphology of pollen development in *O. lata*; and the development of the pollen mother cells will now be taken up in greater detail. It is a noteworthy fact that the mother cells of a given loculus show but little variation in their stage of development. Generally, however, there is a slight progression from top to bottom of a loculus; e.g., when the pollen mother cells at the top of a loculus are in pre-synaptic stages, those at the bottom may be in the late post-synaptic stages of the prophase; or when the mother cells at the top of a loculus are in metaphase of the heterotypic division, those at the bottom may have reached the telophase with reconstructed nuclei. In comparing different loculi of an anther or different anthers of the same flower, the variation is often much greater. Thus in some loculi the pollen mother cells may be in synapsis; while in others of the same or another anther in the same flower, they may be in metaphase of the heterotypic

division. This irregularity is probably connected with the failure in pollen development.

The cytoplasm of the mother cells in the "resting" condition contains numerous large spherical starch grains. In safranin-gentian they stain pale blue and the central hilum is made more prominent by its retaining a highly refractive globule of xylol. These starch grains are thus very conspicuous objects in the mother cells (*fig. 3*). With iodine they appear brown and angular, and the iron-haematoxylin stain leaves them invisible, so that very different appearances of the mother cells are obtained according to the method of treatment. During the prophases of the first mitosis the starch grains become more abundant, until the cytoplasm is literally packed with them. This reserve store of nutriment is used up during the reduction divisions, and no starch grains are to be seen in the pollen tetrads. In iron-haematoxylin the cytoplasm of the pollen mother cells frequently presents a rather obscurely radiate appearance during the pre-synaptic stages. In the earlier or "resting" condition of the nuclei in the mother cells they contain one large nucleolus, which is commonly placed near the nuclear wall and contains one to several vacuoles of varying size with highly refractive walls (*fig. 9*). In addition, two or more smaller chromatic bodies are usually found distributed near the nuclear membrane. They appear connected and often surrounded by the delicate reticulum scattered through the nucleus. The large nucleolus is several times the size of the smaller bodies. Sometimes it is in the center of the nucleus, and the smaller bodies are appressed to the nuclear wall. Vacuoles are usually observed only in the large nucleoli, but occasionally minute ones may be seen in the smaller chromatic bodies. More rarely two large nucleoli of nearly equal size may be found. The small round chromatic masses arise from the more angular chromatic bodies found at the nodes of the reticulum in an earlier stage (*fig. 8*). These bodies probably correspond to Overton's "prochromosomes," though they have not been studied fully. The next stage leading towards the synapsis condition is apparently the beginning of contraction of the nuclear reticulum. The nuclear membrane is often extremely delicate at this time, and as indicated in *fig. 10*, the nucleolus is frequently surrounded by a compact reticulum which has contracted from the nuclear membrane,

and in which are embedded, especially near the margin, the "pro-chromosomes," forming an incomplete circle around the periphery. The material in which are found the beginnings of this contraction, as well as its later stages, was fixed and stained in the same manner as the slightly earlier stages in which it does not occur.

The transition from this stage to the regular spirem stages has not been followed. The pre-synaptic stages with a continuous spirem last for a comparatively long time, as shown by the frequency of their occurrence. The spirem is at first extremely delicate, very long and tangled (*fig. 11*). One large nucleolus is always seen at this time, but one or more smaller ones may also be present. In a later stage the nucleolus is for a time closely appressed and somewhat flattened against the nuclear wall. Loops of the spirem extending to the wall frequently give a false appearance of doubling (*fig. 14*), but though a great many spirem stages have been studied, a double thread has never been observed. The small size of the nuclei, however, and the delicacy of the spirem thread make it very difficult to determine this point.

The spirem gradually contracts into a dense ball with a few loose threads projecting irregularly (*fig. 12*). In this closely contracted condition it may form a body about the size of the nucleolus, which can only be distinguished from the latter by its somewhat irregular outline (*fig. 13*). In the next stage observed the spirem is again loosely arranged in the nuclear cavity, but is greatly contracted in length and several times the thickness of the original spirem before the contraction stage. This is shortly before the spirem breaks up into chromosomes, and the thread is at this time about the same thickness as the chromosomes in the later prophase. Throughout the earlier stages of the prophase the spirem thread is continuous, and, judging from its frequency, the duration of the long delicate spirem previous to the synapsis stage is considerably longer than that of the short thick spirem after synapsis and before it segments into chromosomes. In the latter stage, before segmentation into chromosomes, there is frequently found, besides the spirem, a ring-shaped body of chromatic material exactly like the spirem in thickness and staining power. This has evidently been cut off from the spirem (*figs. 17, 18, 19*). Frequently there are two such rings.

*Fig. 16* probably shows the origin of one of them by the cutting off of a loop of the spirem. There can be no doubt that these bodies arise from the spirem thread previous to the breaking up of the remainder of the thread into chromosomes. The nucleoli at this time are large, globular, and deeply-staining. *Fig. 20* shows a slightly later stage, in the prophase, where two of these ring bodies, which I shall call "heterochromosomes"<sup>3</sup> on account of their later history, are present in addition to the nucleolus and several chromosomes. That these are not the ring-shaped chromosomes frequently found in the prophase of the heterotypic mitosis is shown by their later history; for the rings do not segment in metaphase, forming V- or U-shaped structures, but pass towards the poles of the spindle or into the cytoplasm undivided and retaining their ring form. The shape of the ordinary chromosomes, which are nearly globular or slightly elongated and pear-shaped, precludes the possibility of the pairs forming the X, Y, V, and O shapes which characterize the heterotypic mitosis when the chromosomes are long and rod-shaped.

The heterochromosomes are not always formed, however, and hence if the original pollen mother cells were all alike in the early synapsis stages, two kinds of pollen cells must result; for, as will be shown later, the heterochromosomes probably represent a portion of the chromatin which is rejected and disappears in the cytoplasm, so that the daughter nuclei must differ in their chromatin content. *Fig. 21* shows thirteen chromosomes in the prophase at a later stage, when they are condensed to a considerably smaller size. The adjacent section shows a nucleolus and one chromosome, but no heterochromosomes are present. It has been determined from a number of counts in the prophase that the sporophyte number of chromosomes, exclusive of the heterochromosomes (or at least when the latter are not present), is fourteen. Whether the number is the same when the heterochromosomes are present has not yet been determined with

<sup>3</sup> The term was proposed by MONTGOMERY as a general term for the aberrant chromosomes in Hemiptera, and is used in preference to "idichromosomes," because the latter term has been applied to a certain type of heterochromosomes by WILSON (37). More recently MONTGOMERY (20) has proposed the term "allosome" as a general term for all aberrant chromosomes, and "monosome" for one which passes undivided to one of the daughter nuclei. The latter term would be most suitably applicable to the bodies described here.

certainty, but it appears probable that it is less. BEER (3) states that the reduced number of chromosomes in *O. longiflora* is seven; hence these two species probably agree in the number of their chromosomes. Frequently the chromosomes in the late prophase show evident pairing, but it is usually difficult to represent this accurately in a drawing, because the members of a pair generally lie in different planes. However, indications of such a pairing are shown in figs. 21 and 22. The positions of the chromosomes make it certain that this is an apposition and pairing of whole chromosomes and not a splitting. This is evident, for the chromosomes are completely separated when first seen in prophase and are only arranged in pairs later, on the equatorial plate.

Fig. 22 shows another prophase with ten chromosomes, no heterochromosomes, and a nucleolus in which the stain has been withdrawn from its outer shell except on one side. This condition of the nucleolus is common enough to cause one to suspect that it possesses some significance. Possibly the side of the nucleolus in which the amphipyrenin retains the stain is the one which was earlier attached to the nuclear wall. However I have no observations to prove this. The nucleolus now rapidly loses its staining power, and in the late prophase, after the chromosomes are formed, frequently stains only with orange G in an iron-haematoxylin-orange stain. It gives no indications of loss of shape or size before it disappears, and I think the most probable supposition is that it is quickly dissolved in the cytoplasm when the nuclear membrane breaks down. The evidence is clear that it disappears about this time, and it is very rarely seen as late as metaphase. I may state here that the stages of its reappearance in the reconstructed daughter nuclei at the end of the heterotypic mitosis are in the reverse order of its disappearance. Soon after the nuclear walls appear around the daughter nuclei, when the nuclear sap has increased in quantity, the nucleoli appear (fig. 42) as pale yellow-staining bodies, usually several in number and frequently attached to the nuclear wall. It is impossible to believe that these bodies are reconstituted from fragments of the original nucleoli thrown out into the cytoplasm during the first mitosis and collected again into the daughter nuclei, as described by SCHAFFNER (26) for the megasporre divisions in *Lilium philadelphicum*. For

in the last stage of their disappearance they retain their shape but lose their staining power, as if the deeply staining substance (chromatin?) had been dissolved away, leaving the yellow-staining ground-substance, and in the first stage of their reappearance the yellow-staining material only is present. I think this precludes the possibility of identifying the bodies in the cytoplasm, which stain like chromatin, as fragments of the nucleolus.

#### THE HETEROTYPIC MITOSIS

Thus far the description of events in the pollen mother cell has related only to *O. lata*, because the corresponding stages in the *O. Lamarckiana* side of the hybrid have not been studied. There is every reason to believe, however, as will be shown later, that the history is the same except for the difference in the number of chromosomes, because heterochromosomes are found very commonly in the *O. Lamarckiana* side of the cross, in the metaphase of the heterotypic mitosis, and they doubtless have the same origin as in *O. lata*. These heterochromosomes are the first described in plants, and a discussion of their possible significance will be deferred to the end of the paper.

*Fig. 23* shows a metaphase of the heterotypic mitosis in *O. lata*. There are shown twelve chromosomes (some of which are in pairs of equal size) and a heterochromosome. *Fig. 24* shows two heterochromosomes of unequal size far out in the cytoplasm.

*Figs. 25* and *26* are from sections of spindles showing the heterotypic mitosis in the *O. Lamarckiana* hybrid. In *fig. 25* the heterochromosomes are of equal size and are found on the same end of the spindle. This condition is very commonly found. In *fig. 26* the heterochromosomes are in the cytoplasm and not attached to the spindle; one of them is larger than the other and they are apparently proceeding towards opposite poles of the spindle.

*Fig. 27* is a side view of a spindle showing a small chromosome out of position near the equatorial plate. A similar body is seen in *fig. 26*. What significance these may have is not known. Their early history has not been observed. They are not ring-shaped, like the typical heterochromosomes, but they frequently stain less deeply than the regular chromosomes. These bodies were only

observed a few times, while the heterochromosomes are very common although they are certainly absent from some pollen mother cells.

*Figs. 29, 30, 31, and 32* are sections of the equatorial plate of the *O. Lamarckiana* hybrid, showing one or more heterochromosomes. The chromosomes are so densely grouped at this time that it is often impossible to make out the individuals. This renders some of the figures unsightly, but they are intended to be merely accurate representations of what is seen on the spindle.

After passing into the cytoplasm, where they are generally found in metaphase, the heterochromosomes apparently degenerate and disappear. They gradually become smaller and stain less deeply, though retaining their ring form, as in *fig. 32*. Finally they are so small and inconspicuous as to be scarcely distinguishable from the granules (microsomes) of the cytoplasm. It is probable that they usually disappear before the telophase of the heterotypic mitosis. Several other points in regard to their history have yet to be determined.

*Figs. 34 and 35* are equatorial views. *Fig. 34* shows several chromosomes together with a heterochromosome, and also a very large or giant ring chromosome, which apparently contains enough chromatin for several chromosomes. *Fig. 35* shows another; and these two cases were the only ones observed. They are apparently quite distinct from the heterochromosomes already described, although they are ring-shaped. No other observations have been made in regard to them.

*Fig. 35* is a polar view of the equatorial plate of the heterotypic mitosis in the *O. Lamarckiana* hybrid. The apparently unequal pairs are probably due to foreshortening, owing to one chromosome lying in a lower plane. *Figs. 36 and 37* are early anaphases in the *O. Lamarckiana* hybrid. *Figs. 38 and 39* are late anaphases. *Fig. 38* shows clearly the premature splitting of the chromosomes for the homotypic division. This splitting may occasionally be observed in late metaphase, and in the late anaphase the separation of the halves is often complete. V-shaped chromosomes were not observed, the split apparently taking place simultaneously throughout. Thus it is clear that the heterotypic mitosis distributes whole chromosomes, which were arranged in pairs (though not at first in contact) in the prophase; while the homotypic mitosis is a typical division, separating

the halves of the chromosomes already formed by splitting in the anaphases of the heterotypic division. The heterotypic is thus the reducing division. The shape of the chromosomes of course makes it impossible to determine whether the second splitting is longitudinal or transverse.

*Fig. 40* shows a telophase in which the reconstruction of the daughter nuclei has begun. Delicate nuclear walls are appearing and the chromosomes have assumed angular, lobed, and irregular forms, but very little nuclear sap has yet appeared. Materials staining like chromatin are scattered through the cytoplasm. *Fig. 41* shows the same stage, in which bodies as large as chromosomes and taking the same stain are scattered through the cytoplasm (particularly in the region of the daughter nucleus). It has already been shown, I think, that these bodies cannot be fragments of the nucleolus. Neither are they likely to be fat bodies, since the cytoplasm in earlier stages contained abundant storage nutritive materials in the form of starch. It seems then most reasonable to conclude that these bodies are actually chromosomes, and fragments of chromosomes which for some reason have become separated from their fellows on their way to the poles of the spindle. It is difficult to understand, however, what forces carry them into the cytoplasm, for among hundreds of spindles studied no irregularities in their structure have been observed which would cause the chromosomes to be distributed irregularly. Indeed the heterotypic spindle is notably regular in form. The earlier stages of spindle formation have not been studied, however, and it may be that these would account for some of the chromosomes not being on the spindle. It is doubtful if the heterochromosomes are ever attached to the spindle. Of course the position of chromosomes near the nuclear membrane is easily accounted for by their having merely failed to pass to the poles of the spindle, but this will not explain the position of many which are found scattered in the cytoplasm throughout the cell and frequently near its periphery.

*Fig. 42* is a polar view of a daughter nucleus in a later stage of reconstruction. The nucleus has grown greatly in size, owing to a very rapid increase in the nuclear sap. At the same time the chromosomes have become drawn out into irregular shapes, are much reduced in size and stain less deeply. Several nucleoli have appeared, but

they do not take the chromatin stain. In an iron-haematoxylin-orange stain they take only the orange; thus it is difficult to see how the view that the nucleoli of the daughter nuclei are reconstructed from fragments of the former nucleolus (SCHAFFNER 26) can apply here, for fragmentation of the nucleoli before their disappearance has never been observed, and their disappearance is preceded by loss of staining power, as if the chromatic-staining substance were first dissolved away, leaving the pale ground-substance. The method of final disappearance of the latter has not been observed. As already mentioned, the reverse series of events occurs in the reappearance of the nucleoli in the daughter nuclei. When first observed in the daughter nuclei, they have their characteristic size and shape, but stain yellow and only acquire the chromatic stain later. They were never observed until there was a considerable accumulation of nuclear sap, and are certainly not visible until some time after the membranes of the daughter nuclei have been formed. These changes in the staining power of the nucleoli might be attributed to chemical changes in the nuclear sap, the medium in which they float, such as would alter the staining reaction of bodies contained in it. But while the nucleolus undergoes these changes before its disappearance, the chromosomes remain constant in their staining properties, which makes it appear more probable that the change in staining is due to actual loss of material from the nucleolus.

Several of the figures (figs. 37, 38, 39) show that the number of chromosomes in the *O. Lamarckiana* hybrid is greater than in *O. lata*. A count has not been made in the prophase, but from numerous counts in the metaphases and anaphases, the sporophyte number is found to be at least twenty and probably very near that number (fig. 37). It will be remembered that the sporophyte number in *O. lata* is fourteen. The bearing of these facts will be discussed later.

The study of several other interesting features of the telophase of the heterotypic as well as of the homotypic division has not yet been completed.

#### The pollen tetrads

The pollen tetrads in *O. lata* show numerous interesting features. As already stated, development to this stage frequently takes place,

though not always. When development takes place, the tapetum is also found to be normally developed and functional. In some cases, as in *figs.* 43 and 44, the normal number of four nuclei is found (three shown in the figures), though these may vary in size (*fig.* 43) and in chromatin content. In other cases (*fig.* 45) chromatic bodies are found in the cytoplasm. In *fig.* 46 one of the nuclei has two others lying near it. *Fig.* 47 shows a tetrad with extra nuclei. Occasionally large and irregular shaped nuclei are found, as in *fig.* 51. *Fig.* 48 is a section of a tetrad showing an extra nucleus formed from a single chromosome and connected by spindle fibers with one of the larger nuclei. *Fig.* 49 is a portion of a tetrad showing a nucleus with a smaller one beside it; and *fig.* 50 similarly shows two small extra nuclei, each containing two chromosomes. Pairing of the chromosomes in the large nuclei of the tetrad is sometimes seen. Chromosomes are frequently found just outside the nuclear wall; this is probably due to tardiness in passing to the poles of the spindle. Such chromosomes may or may not be surrounded by a "nuclear membrane." Most of the pollen tetrads show evidences of breaking down, and a great many are more or less completely degenerated. In the *O. Lamarckiana* hybrid irregular-shaped pollen grains are quite common in some anthers; *fig.* 52 shows some of these irregularities. There may be extra lobes or fewer lobes than the normal, and they may be irregularly placed. Occasionally complete sterility of certain anthers of a flower may be found in the *O. Lamarckiana* hybrid, as in *O. lata*.

#### Discussion

A full discussion of the literature bearing on the facts set forth in this paper will not be attempted at this time, as the work touches upon several distinct fields. But I shall mention a few of the most important papers bearing directly upon these investigations, and close with a short discussion of the general bearing of the most important results.

#### THE EXTRA NUCLEI IN POLLEN TETRADS

WILLE published a paper (34) on pollen development in the angiosperms in 1886, before botany had acquired its present technique, in which he cites a list of cases of extra nuclei in the tetrad

divisions. He remarks that these are chiefly cultivated plants, and suggests that the extra nuclei are indications of greater variability in structure resulting from cultivation. This was of course before the modern views of chromosome reduction and alteration of generations had been developed. The fact that extra nuclei are so commonly found among the irregularities in the divisions of the pollen mother cells in hybrids, together with the well-known fact that cultivated plants are very commonly hybridized, would seem to lay open to suspicion the purity of any species in which such irregularities occur. Even *Hemerocallis fulva*, which is commonly quoted as a pure species having these irregularities, is well known to have been long under cultivation, and three varieties of it are described by BAILEY (2). The other members of the list given by WILLE (*l. c.* 60), with mention of their cultivation and varieties, taken from BAILEY's *Cyclopedia of American Horticulture*, are given below.

Name	Varieties	
<i>Ampelopsis hederacea</i>		
— <i>A. quinquefolia</i> . . . . .	..	cult. climbing vine
<i>Azalea indica</i> . . . . .	..	cult. and hybridized
<i>Begonia</i> sp. . . . .	..	cult. and variable
<i>Charitania ilicifolia</i> . . . . .	..	not in Bailey
<i>Cornus sanguineus</i> . . . . .	2	cult. shrubs
<i>Elatine hexandra</i> . . . . .	..	not in Bailey
<i>Ficaria ranunculoides</i> . . . . .	..	not in Bailey
<i>Fuchsia</i> sp. . . . .	..	cult., very variable, and probably hybridized
<i>Funkia ovata</i> . . . . .	14	cult.
<i>Hemerocallis fulva</i> . . . . .	3	cult.
<i>Lonicera coerulea</i> . . . . .	..	cult. shrubs
<i>Prunus cerasus</i> . . . . .	..	cult. and hybridized
<i>Rumex patientia</i> . . . . .	..	cult. and nat.
<i>Scleranthus annuus</i> . . . . .	..	not in Bailey
<i>Stellaria glauca</i> . . . . .	..	not in Bailey
<i>Sympodium officinale</i> . . . . .	1	cult.
<i>Syringa persica</i> . . . . .	2	cult.

An examination of the list shows that all but five are forms cultivated in America, and most of these are either open to suspicion as hybrids or known to have been hybridized. In the light of these facts, a careful examination of the history of all forms in which such extra nuclei are known to occur would be advantageous. It would

\* Two other varieties are given by J. G. BAKER in "A revision of the species of herbaceous gamophyllous Liliaceae" in *Jour. Linn. Soc.* 11:366. 1871.

thus be possible after a time to decide whether they result from hybridization only, or whether they may be the result of some other constitutional derangement caused by cultivation. It should be mentioned in this connection that Miss LYON (16) described five and six spores in a mother cell of *Euphorbia corollata*, which is a common species in this region. In this case the daughter cells are stated to be all of the same size, however, which suggests that the number may have been produced by extra divisions of one or more of the daughter cells, rather than by some of the chromosomes becoming scattered in the cytoplasm. The same explanation may apply to some of the other forms mentioned; and it is also necessary to assume extra divisions in cases such as *Lonicera caerulea*, where as many as fourteen daughter cells are figured in one mother cell. FULLMER (7) thought that such extra mitoses would account for most of the conditions in *Hemerocallis*, but in this he was probably incorrect.

A more careful study of some of these forms, and of some of those said to produce only one, two, or three pollen grains from a mother cell, as described in the paper of JUEL (14) on *Carex*, would doubtless furnish many interesting results and aid in a determination of the causes of these irregularities.

#### THE PHENOMENA OF POLLEN DEGENERATION

During the last few years a number of cases of sterility in the pollen of hybrid plants have been described, with interesting morphological and cytological results. JUEL (13) in 1900 described pollen development in the sterile *Syringa Rothomagensis*, which is a cross between *S. vulgaris* and *S. persica*. He also studied the corresponding stages in the parents for comparison. In *S. vulgaris* only about 50 per cent. of the pollen grains were found to be normal, and in *S. persica* good grains were few. Practically the same conditions of development were found in *S. persica* as in the hybrid. A great variety of irregularities in the reduction divisions was found in the hybrid. As in *O. lata*, the degeneration sometimes began as early as the synapsis stage, but generally the tetrad divisions were completed. A drawing apart of the nucleus into two may occur in the spirem stage or after the spirem has segmented into chromosomes. The latter at least could not be called amitosis. In the first reduction

mitosis chromatin appears in the cytoplasm, as in the *Oenothera* hybrids studied. The spindle is also sometimes multipolar and irregular. Several nuclei may be found in a tetrad, and also chromatin scattered in the cytoplasm, as in *O. lata*. This hybrid shows a greater variety of irregularities than have been observed in any other hybrid described.

GUYER (10, 11) found in the spermatogenesis of hybrid pigeons similar conditions with extra and irregular spindles and incomplete pairing of the chromosomes. CANNON (5) found in hybrid cotton, *Gossypium barbadense*  $\times$  *G. herbaceum*, that degeneration of the pollen mother cells sometimes begins in the resting stage previous to synapsis. The divisions in some of the pollen mother cells are amitotic, the nucleolus dividing first, followed by equal or unequal segmentation of the nucleus and its reticulum. In the case of equal segmentation, pollen tetrads of normal appearance may be formed. Multipolar spindles, with many small nuclei, were also found. In fertile hybrids of certain races of peas, which obey Mendel's law, CANNON (4) found no difference in pollen development between the hybrids and their parents. He states that there is a pairing of the chromosomes in the telophase of the last sporogenous division previous to synapsis. It should be said, however, that these two papers are not as complete and satisfactory as some of the others.

ROSENBERG'S (23, 24) study of pollen development in *Drosera longifolia obovata*, a hybrid of the intermediate type, between *D. rotundifolia* and *D. longifolia*, but nearer *D. longifolia*, shows that some of the smaller chromosomes of *D. longifolia* are left behind in the cytoplasm in the first and second divisions, where they also form small nuclei. Later the greater number of the pollen grains lose their contents. The embryo sac development also usually stops at the binucleate stage. The sterile hybrid sweet peas (*Lathyrus odoratus*) studied by GREGORY (9) show an earlier degeneration of pollen than the other cases cited. Portions of the spirem are often destitute of chromatin and the chromosomes frequently are pale, which GREGORY attributes to lack of nutrition. The nucleolus, he thinks, serves to store most of the chromatin between the reduction mitoses. Other conditions simulating amitosis in the pollen mother cells are illustrated. Degeneration of the pollen mother cells usually takes place

in the prophase, but occasionally the first spindle is formed, though it is irregular. Development never proceeds to a later stage. The sterility is here a Mendelian character, recessive and correlated with a certain somatic character, the two always appearing together.

TISCHLER (31) ascribes the sterility of the hybrids of *Ribes* and *Bryonia*, as well as those of *Lathyrus odoratus*, to the influence of long culture. He studied three hybrids of *Ribes*: (1) *R. intermedium* = *R. sanguineum* × *R. nigrum*; this hybrid is fertile, producing 10–15 per cent. good pollen grains; (2) *R. Gordonianum* = *R. aureum* × *R. sanguineum*; absolutely sterile and showing great vegetative luxuriance; (3) *R. Schneideri* = *R. grossularia* × *R. nigrum*; sterile. He finds a splitting of the spirem in the strepsinema stage, and that the chromosomes when formed lie in pairs in the prophase. As in *O. lata*, the heterotypic is a reduction division, the homotypic an ordinary equation division, and the chromosomes split (longitudinally) in the anaphase of the first mitosis. In all the hybrids, as well as in the parents, e. g., *R. sanguineum*, the reduction divisions are mostly normal, though in *R. Gordonianum* occasional extra spindles are found in the cytoplasm; otherwise its tetrad divisions cannot be distinguished from those of *R. intermedium*; but the sterile grains do not grow in size. The first evidences of degeneration appear then in *Ribes* later than in any of the others mentioned, i. e., usually after the reduction divisions are completed.

In a sterile hybrid of *Begonia alba* ♀ × *B. dioica* ♂, TISCHLER (32) finds that when the reduction divisions in the pollen mother cells are irregular, they are invariably preceded by abnormalities in synapsis; but that when synapsis is accomplished regularly the reduction divisions are also carried through without irregularities. There is often unequal division of the chromosomes on the heterotypic spindle. In the tetrad divisions, chromosomes in the cytoplasm and extra nuclei are relatively seldom found. The pollen grains are very often degenerate and of unequal size. In the female gametophyte the megasporangium mother cells divide normally, but the embryo-sac mother cell degenerates. Whether another embryo-sac is formed later from the sporophyte tissue, as shown by TISCHLER for *Cytisus Adami* (29), *Ribes Gordonianum*, and *Syringa chinensis* (30), has not been determined.

In some of the papers sufficient attention has not been given to a comparison of the adult characters of the hybrid with those of the parents, *i. e.*, whether the hybrid is a mosaic, Mendelian, an intermediate, or goneoclinic to one parent, constant and invariable or breaking up into different types, etc. It would be particularly valuable to know in every case to what particular type the hybrid whose germ cells are studied conforms, and what variations if any occur.

#### THE POSSIBLE CAUSES OF STERILITY IN HYBRIDS

That hybrids are frequently sterile has of course long been known and commented upon. The recent morphological and cytological papers already cited have not reached an explanation of its cause, though various suggestions have been made. Some of the causes suggested may be classified into several general categories which are not, however, necessarily mutually exclusive: (1) lack of "nutrition" in the parts affected, causing, *e. g.*, failure in development of the tapetum or low staining capacity of the chromatin in the germ cells; (2) the influence of long culture (whatever that may be); (3) irregularities of development in the germ cells, appearing during synapsis or the reduction divisions and due to some lack of harmony (variously expressed) between the idioplasts; (4) some more deep-seated phenomenon affecting the physiology of the whole plant. This is not an attempt to classify all the possible causes of sterility suggested, but it will be seen that the factors are stated from widely differing points of view and with varying amounts of definiteness. Some of them might be viewed as effects rather than as causes.

In *O. lata*, if the tapetal cells always showed a degeneration before the pollen mother cells, we might conclude that the failure of the tapetum to function (in the secretion of nutrient for the pollen grains, which is generally conceded to be its chief function) was the immediate cause of the abortion of the pollen. But this is not always the case; for degeneration in the pollen mother cells may appear as early as in the tapetal cells or earlier; while GREGORY states that in the hybrids of *Lathyrus odoratus* the "somatic cells" are normal throughout. GUYER (10) suggested in 1900 that incompatibility of the chromosomes or plasms, as shown in the prophases of the first reduction division, is the source of the subsequent irregularities in

spermatogenesis, and consequently the cause of sterility in hybrid pigeons. But in the case of *O. lata* such an explanation can scarcely suffice, for we cannot imagine that any incompatibility or failure of fusion of the maternal and paternal plasms or chromosomes could almost simultaneously bring about the disintegration of the tapetal cells. GREGORY (9) suggests that "it seems more likely that sterility of the male organs is the expression of some deeper-lying phenomenon affecting the whole plant," and a similar view is held by TISCHLER (31). Thus it appears that the cause must be sought in some more wide-spread phenomenon in the hybrid, causing general lack of nutrition of the parts which degenerate; hence the tapetum and pollen mother cells degenerate simultaneously and for the same reason. Recent papers, which need not be referred to in this connection, have also shown that hybridism may cause abnormalities or failure of development in the female gametophyte, though this is apparently less common. These facts, together with the usually normal and in some cases increased luxuriance of the sporophyte, point to the gametophyte as that part of the life history which suffers chiefly from "lack of nutrition." The sporophyte generation can evidently be carried through successfully if fertilization is once accomplished; but something which first displays itself as lack of nutrition (as shown by the failure of the tapetum to function, as well as perhaps by the pale staining of the chromatin of the pollen mother cells in some cases) prevents the normal development of the gametophyte.

That this is not the whole case, however, is shown by the fact that the reduction divisions are abnormal, even when the tapetum appears perfectly developed and functional. Hence something other than "lack of nutrition" must be the cause of these abnormalities in development. The ingrowth of the median wall layers to fill the loculus after the disappearance of the pollen, points rather in the opposite direction. But perhaps a harmony of these two points of view may be reached. If the view, recently emphasized by COOK and SWINGLE (6a), ALLEN (1), CARDIFF (6), and others among botanists, may be accepted, namely, that the maternal and paternal chromatin, which contribute to the fusion nucleus at fertilization, maintain their separate identity throughout the sporophyte generation, so that the act of fertilization is really only completed with the fusion of the

spirem threads in synapsis, as a preliminary to sporogenesis; then, as these authors point out, the synapsis stage becomes the critical point in the life cycle of the individual. It is conceivable that the maternal and paternal chromatin after entering the same nucleus in fertilization could continue in this condition throughout the sporophyte history without interfering with development, but that the intimate union which occurs in synapsis, where an interchange of material between the parental idioplasms probably takes place, might lead to the development of "incompatibilities" between the plasmas, such as are exhibited in irregularities and disturbances in the reduction divisions, and finally more or less complete failure of later development. It may be remembered in this connection that in *O. lata* no irregularities in the prophases were observed until after the contracted condition of the spirem.

This discussion is intended to serve merely as a partial analysis of the problem, of which it does not offer a solution. A larger body of facts is very desirable, correlating cytological studies of hybrids with observations of their variations and their relations to the parental forms in all their characters. It may then be possible to determine whether observable differences in the mechanism of inheritance as exhibited by the chromosomes occur in Mendelian, intermediate, variable, and other types of hybrids.

#### THE CHROMOSOMES AND MUTATION

It is believed that one of the most important discoveries connected with the investigation thus far has been the observation of certain chromatic bodies with a peculiar origin and history, which have been designated heterochromosomes. Since they have been found in the dominant *Lamarckiana*  $\times$  *lata* hybrid, I believe it is probable that they will also appear in *O. Lamarckiana* itself. This point is to be investigated as soon as possible. If this surmise is found to be true, then the pollen mother cells in which these bodies occur must differ in chromatin content, and hence probably in hereditary value, from those in which they are absent, provided that pollen grains from both these classes of mother cells reach maturity. It is highly probable that the latter supposition is true in the *O. Lamarckiana* hybrid, for an abundant amount of pollen reaches maturity, so that in crossing

experiments fertilization is as easily accomplished as with pure *O. Lamarckiana* pollen; which increases the probability that no extensive deterioration of pollen grains occurs. POHL (22) states, however, that only about 70 per cent. of the pollen grains of *O. Lamarckiana* are perfect. Whether the deterioration is selective is of course not known.

The fact that the heterochromosomes originate from the spirem by the cutting off of a portion of its length after synapsis, and later degenerate, suggests that in this manner a portion of the chromatin is dispensed with in certain pollen mother cells, while in others where heterochromosomes do not occur it is all retained. Those who hold strictly to the individuality of the chromosomes, and that they represent in some way certain specific characters or groups of characters, would probably maintain that by this means the specific characters represented by the discarded chromatin are lost. LILLIE (15) expresses the view (which he strongly substantiates) that the chromosomes can represent only general characters belonging to the organism as a whole; such as color, stature, pubescence, etc. These would appear to correspond with the unit characters of DEVRIES, at least in many essentials, for instances of the latter as cited by DEVRIES in the Oenotheras are pigment (*in O. rubrinervis*), stature (*O. nanella* is a dwarf), and pubescence. This loss of chromatin in the germ cells might then appear to be a method of dispensing with certain unit characters of the organism. A great amount of additional work will be required, however, in the study of somatic characters in connection with chromosome morphology, before it will be possible to correlate these two categories of structure, if such a correlation exists.

ROSENBERG (25) has recently published a further paper on the Drosera hybrid, in which he finds that occasionally two pollen grains of a tetrad have the characters of one parent and two those of the other parent, the supposition being that this is preceded by segregation of the maternal and paternal chromosomes in the first reduction division. This may be considered a proof that the segregation of parental characters when it occurs takes place during the reduction divisions. In the light of such facts it seems highly probable that mutations in the Oenotheras will be found to originate during the

reduction mitoses, and perhaps from irregularities in the distribution of chromosomes. The results of the paper mentioned above would also appear to show that a relation exists between chromosome morphology and the morphology of the pollen grains.

The origin and early stages of some of the bodies classed as "idiochromosomes" by WILSON (36) are similar to those I have found in Oenothera. In *Lygaeus turcicus*, one of the Hemiptera, a large and a small idiochromosome are found in the prophase, the large one first appearing about the time of synapsis. Slightly later it is seen as a more deeply staining longitudinally split thread among the chromosomes, which are also thread-like bodies at this time. During this period a smaller, somewhat elongated idiochromosome is also present. The larger idiochromosome condenses to a globular shape and is for a time attached to the plasmosome, but retains a central cavity, which represents the original longitudinal split. This makes its appearance quite similar to those described in Oenothera. The later history of the idiochromosomes in Lygaeus and other Hemiptera however, is quite different from those of Oenothera, and need not be taken up here. Small chromosomes, which possibly correspond with WILSON's *m*-chromosomes, are also occasionally found in Oenothera (figs. 26, 27).

Miss STEVENS (28) has described chromatic bodies in the insects which correspond with the heterochromosomes of Oenothera in parts of their history. In *Stenopelmatus*, the "sand-cricket," a chromatic "element *x*" appears in synapsis in the spermatocytes attached to an end of the spirem. It passes undivided into one of the spermatocytes of the second order and there degenerates during the resting stage between the maturation mitoses. Miss STEVENS believes this to be rejected chromatin, analogous to that observed in the ovogenesis of many forms. In the animal *Sagitta*, which is hermaphrodite, Miss STEVENS (27) found in spermatogenesis bodies which resemble the heterochromosomes of Oenothera in some respects. This "element *x*" appears in the spermatogonial divisions, where it divides before each mitosis. It also divides in the prophase of the first maturation division, and when the chromosomes are on the equatorial plate in metaphase these bodies are found in various

positions on the spindle. But they enter the daughter nuclei and divide again before the second spindle forms, and one of the products thus enters each spermatid. No such body is found in ovogenesis.

The only previous suggestion of heterochromosomes in plants is in a recent paper of CARDIFF (6), in which he mentions a heterotrophic chromosome in the heterotypic mitoses of *Salomonia biflora*. He believes that this chromosome passes to one pole undivided in the first mitosis, but has not studied it further.

The difference found in the number of chromosomes in the mutants of *Oenothera* very strongly favors considering these forms of "specific" rank. I think it will be evident to any one studying carefully and comparing the different mutants that they are quite as distinct and easily distinguishable as are the species of any ordinary genus. The differences in the number of chromosomes is still further and, I think, conclusive evidence that the forms concerned are distinct "species."

It seems necessary to conclude, therefore, that the phenomena of mutation as described by DEVRIES in the genus *Oenothera* are either due to *O. Lamarckiana* being some peculiar type of hybrid in which the earlier crosses are appearing again in comparatively rare numbers; or on the other hand that some process of differentiation, the most probable seat of which is the germ plasm, has led to the production of distinct types of germ cells differing in chromosome morphology and in hereditary value. The middle ground, the assumption that these various mutants are merely the widely fluctuating variations of the original *O. Lamarckiana*, is believed to be no longer tenable.

Moreover, there are several lines of evidence, partly negative, which tend in various ways to discredit the possible hybrid origin of these forms. Some of these arguments will be mentioned.

1. It has been clearly shown by MACDOUGAL (17, 18) and a number of collaborators that *O. Lamarckiana* was originally native to various parts of North America.

2. If the mutants always existed together with the parent form, it seems probable that some at least would have been recognized as species and preserved in European or American herbaria.

3. *O. biennis* and *O. grandiflora*, American members of the genus, have been shown by MACDOUGAL to be mutating.

4. Among all the known types of hybrids, with their widely varying methods of transmission and of variation, none are known, so far as the writer is aware, which present a series of phenomena of variation resembling or comparable to those found in *O. Lamarckiana* and its mutants.

5. Hybrids from crosses among the mutants of *Oenothera* follow the ordinary laws of hybridization; e. g., *O. lata*  $\times$  *O. Lamarckiana* is a Mendelian or dichotomous hybrid; and *O. nanella*  $\times$  *O. rubrinervis* is a hybrid of intermediate type, which breeds true.

6. If *O. Lamarckiana* has itself been hybridized, where did the forms originate with which, on this assumption, it was hybridized? No scrap of evidence that they ever originated from any source except from *O. Lamarckiana* seed has been found. We must then assume that this is their only method of origin until the contrary is proved to be the case.

#### Summary<sup>5</sup>

1. Failure of pollen development in *O. lata* is not due to ingrowth of the tapetum to fill the loculus, as described by POHL, but to some other agency at work in the hybrid, the nature of which is not fully explained.

2. Pollen development in *O. lata* may proceed to the formation of tetrads, but degeneration of both mother cells and tapetum frequently begins in the resting stage or during the prophase of the first mitosis.

3. Heterochromosomes arise in *O. lata* in the prophase after synapsis. There may be one or two such bodies, which are formed as large rings by the cutting off of a portion of the spirem thread before the remainder breaks up into chromosomes. These bodies later are found in metaphase of the heterotypic mitosis, usually in the cytoplasm near the spindle. They do not divide, but become smaller and

<sup>5</sup>As this paper is passing through the press, conditions are being found in the pure *O. Lamarckiana* which will modify the interpretation of the bodies called heterochromosomes. The inference that *O. Lamarckiana* itself has the same number of chromosomes as the dominant *O. Lamarckiana* hybrid is also apparently not borne out by the facts. There seem to be variations in the number of chromosomes, which will require further careful study before an interpretation will be possible.

probably disappear at the end of the first mitosis. These bodies are also found in the *O. Lamarckiana* hybrid, in which they doubtless have the same origin. They probably represent discarded chromosomes, and this is perhaps a means of lessening the number of chromosomes in certain germ cells of the species. Some mother cells do not contain them. In such cells the (sporophyte) count of chromosomes in *O. lata* is fourteen and in the *O. Lamarckiana* hybrid probably twenty.

4. The occurrence of extra nuclei formed by chromosomes left behind in the cytoplasm in the tetrad divisions of the hybrid *O. lata*, as well as in a number of other hybrids recently described, puts under suspicion the purity of any other plant, such as *Hemerocallis fulva*, in which such extra nuclei are known to occur.

5. The inference seems justifiable that the mutations of *O. Lamarckiana* arise during the reduction divisions and that pollen grains which will give rise to mutants differ in their potentialities and probably also in chromatin morphology from the ordinary pollen grains of the plant.

These facts lead to the conclusion that a cytological basis may be found for the phenomena of mutation as observed in the Oenotheras.

THE UNIVERSITY OF CHICAGO

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#### EXPLANATION OF PLATES II-IV

All the figures were drawn with the aid of a Bausch and Lomb camera lucida. A Zeiss 2<sup>mm</sup> apochromat oil immersion lens was used, with Zeiss compensating oculars. The figures are all reduced one-half.

#### PLATE II

##### *Oenothera lata* (dominant)

FIG. 1. Transverse section of a loculus of an anther; pollen mother cells in "resting" stage.  $\times 325$ .

FIG. 2. Slightly later stage; pollen mother cells passing from "resting" condition to early prophase; tapetal cells uninucleate.  $\times 690$ .

FIG. 3. Later stage; pollen mother cells degenerating; tapetal cells binucleate, some of them degenerating.  $\times 690$ .

FIG. 4. Pollen mother cell degenerating; the nucleus has disappeared and irregular deeply staining bodies appear in the cytoplasm.  $\times 1500$ .

FIG. 5. A loculus after the pollen mother cells have disappeared; the remnants of the tapetum form a thin layer partly lining the cavity; the ingrowth of the middle layers has begun.  $\times 325$ .

FIG. 6. A later stage; the middle layers have grown in, almost completely obliterating the loculus.  $\times 325$ .

FIG. 7. Late stage in pollen development; the middle layers are being pressed together; the tapetal cells are narrow radially; the pollen grains are very irregular in shape.  $\times 155$ .

FIG. 8. "Resting" nucleus of pollen mother cell; the nucleolus contains several highly refractive vacuoles; at the nodes of the reticulum are chromatic bodies, probably representing the "prochromosomes" of OVERTON.  $\times 2125$ .

FIG. 9. Slightly later stage; the chromatic bodies are rounded in shape; safranin-gentian;  $3.3 \mu$ .  $\times 2125$ .

FIG. 10. Slightly later stage; the reticulum is beginning to contract away from the nuclear membrane, which at this time is extremely delicate;  $3.3 \mu$ .  $\times 2125$ .

FIG. 11. Spirem stage just previous to synapsis; four nucleoli are present; the nuclear membrane is extremely delicate; the spirem thread is very thin, about half its thickness subsequent to synapsis; Safranin-gentian;  $4 \mu$ .  $\times 2125$ .

FIG. 12. Spirem thread contracted in synapsis; iron-haematoxylin-orange;  $5 \mu$ .  $\times 2125$ .

FIG. 13. Mother cell in synapsis; chromatic masses are scattered through the cytoplasm; iron-haematoxylin-orange;  $7.5 \mu$ .  $\times 2125$ .

FIG. 14. Just after synapsis; loops of the spirem pass outwards to the wall, giving a false appearance of doubling; the nucleolus is flattened and appressed to the nuclear wall, which is more definite at this time; iron-haematoxylin-orange;  $5 \mu$ .  $\times 2125$ .

### PLATE III

Figs. 15-24. *O. lata* (dominant).

FIG. 15. Nucleus at the time of synapsis; the nuclear membrane is very thick and irregularly indented; safranin-gentian;  $4 \mu$ .  $\times 2125$ .

FIG. 16. Portion of spirem after synapsis; it is about double its former thickness and very much shorter; the loop (*a*) of the spirem may indicate the method of origin of a heterochromosome; safranin-gentian;  $4 \mu$ .  $\times 2125$ .

FIG. 17. Slightly later stage showing a portion of the spirem after synapsis; the large ring body, which has been detached from the spirem, is a heterochromosome (*h*); the nucleolus is present and the nuclear membrane is becoming delicate previous to breaking down; safranin-gentian;  $3.3 \mu$ .  $\times 2125$ .

FIG. 18. Section of nucleus showing two nucleoli and a heterochromosome (*h*); the adjacent section contains the spirem; safranin-gentian;  $4 \mu$ .  $\times 2125$ .

FIG. 19. Similar section showing two heterochromosomes (*h*) and the nucleolus; safranin-gentian;  $4 \mu$ .  $\times 2125$ .

FIG. 20. Late prophase, showing nucleolus, two heterochromosomes (rings), and seven other chromosomes with indications of pairing; safranin-gentian;  $3.3 \mu. \times 1500$ .

FIG. 21. Later prophase showing thirteen chromosomes; the nucleolus and one chromosome on adjacent section; no heterochromosomes present; indications of pairing of the chromosomes; the cytoplasm is pushing into the nuclear cavity; iron-haematoxylin-orange;  $7.5 \mu. \times 1500$ .

FIG. 22. Nucleolus and ten chromosomes; the stain is withdrawn from the shell of the nucleolus, except on one side; iron-haematoxylin-orange;  $7.5 \mu. \times 1500$ .

FIG. 23. Metaphase of heterotypic mitosis, showing twelve chromosomes (some paired) and a heterochromosome (*h*);  $5 \mu. \times 1500$ .

FIG. 24. Same stage as last, showing two heterochromosomes (*h*) in the cytoplasm, and ten chromosomes on the spindle;  $5 \mu. \times 1500$ .

FIGS. 25-34. *O. Lamarckiana* (dominant).

FIG. 25. Section of heterotypic spindle, showing two heterochromosomes (*h*) of equal size on the same end of the spindle;  $7.5 \mu. \times 1500$ .

FIG. 26. Same stage, showing chromosomes closely grouped in the equatorial plate; two heterochromosomes (*h*) of unequal size are present, and a very small chromosome (*m*) out of position on the spindle; iron-haematoxylin-orange;  $7.5 \mu. \times 1500$ .

FIG. 27. Showing chromosomes closely grouped in the equatorial region of the spindle, and a small chromosome (*m*) nearer the pole; iron-haematoxylin-orange;  $7.5 \mu. \times 1500$ .

FIG. 28. Showing two smaller paired chromosomes (*m*) at the side of the spindle;  $7.5 \mu. \times 1500$ .

FIG. 29. Polar view of equatorial plate of heterotypic mitosis; one heterochromosome (*h*) present;  $7.5 \mu. \times 1500$ . Fig. 32 is the adjacent section of this nucleus.

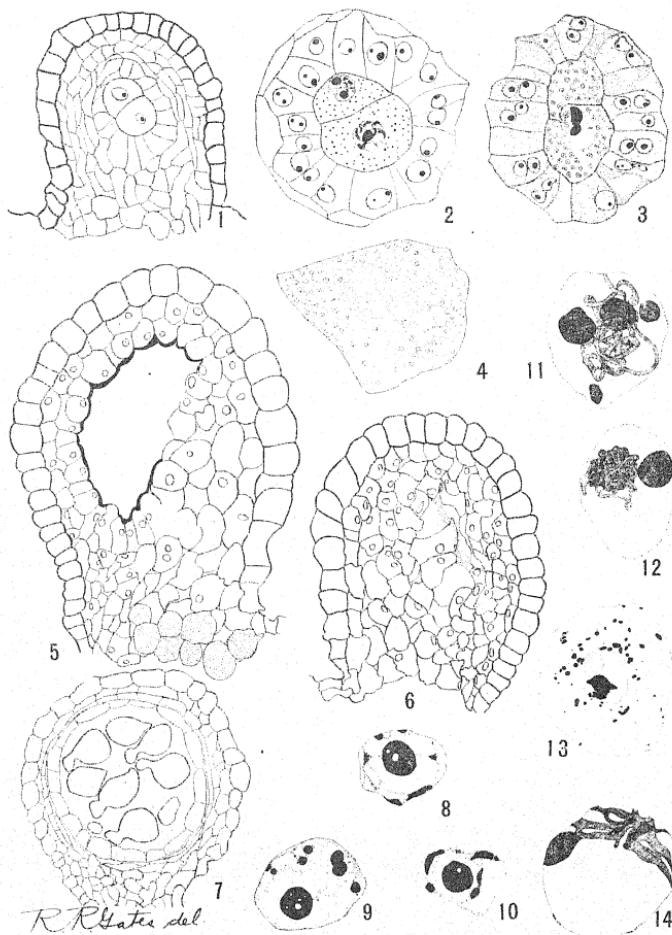
FIG. 30. Section of heterotypic spindle, showing a heterochromosome (*h*), an unequal pair of chromosomes, and an equal pair at the periphery of the cell;  $7.5 \mu. \times 1500$ .

FIG. 31. Portion of the spindle, showing two chromosome dyads and a heterochromosome (*h*);  $7.5 \mu. \times 1500$ .

FIG. 32. Portion of the equatorial plate, showing two very small bodies which stain less deeply than the chromosomes and are probably remnants of a heterochromosome; iron-haematoxylin-orange;  $7.5 \mu. \times 1500$ .

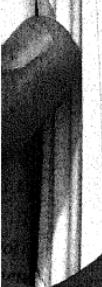
FIG. 33. Section of equatorial plate of heterotypic mitosis, showing a number of chromosomes, a heterochromosome (*h*), and a giant chromosome (*g*), which is very large and ring-shaped;  $7.5 \mu. \times 1500$ .

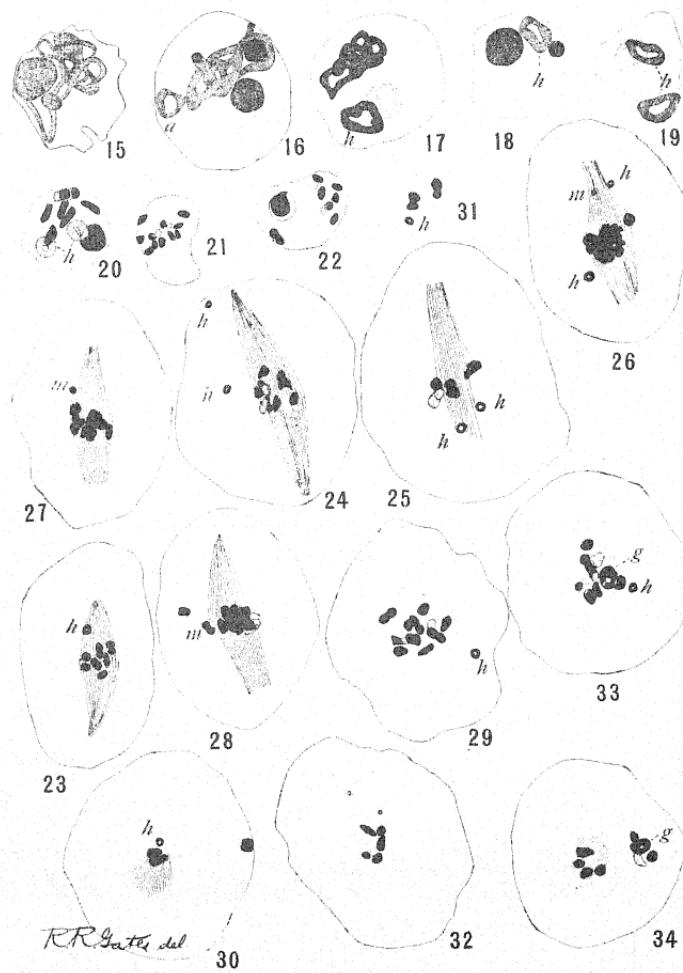
FIG. 34. Equatorial view of heterotypic spindle; the chromosomes at the side are displaced in the figure for the sake of clearness; one of them is the giant chromosome (*g*);  $7.5 \mu. \times 1500$ .



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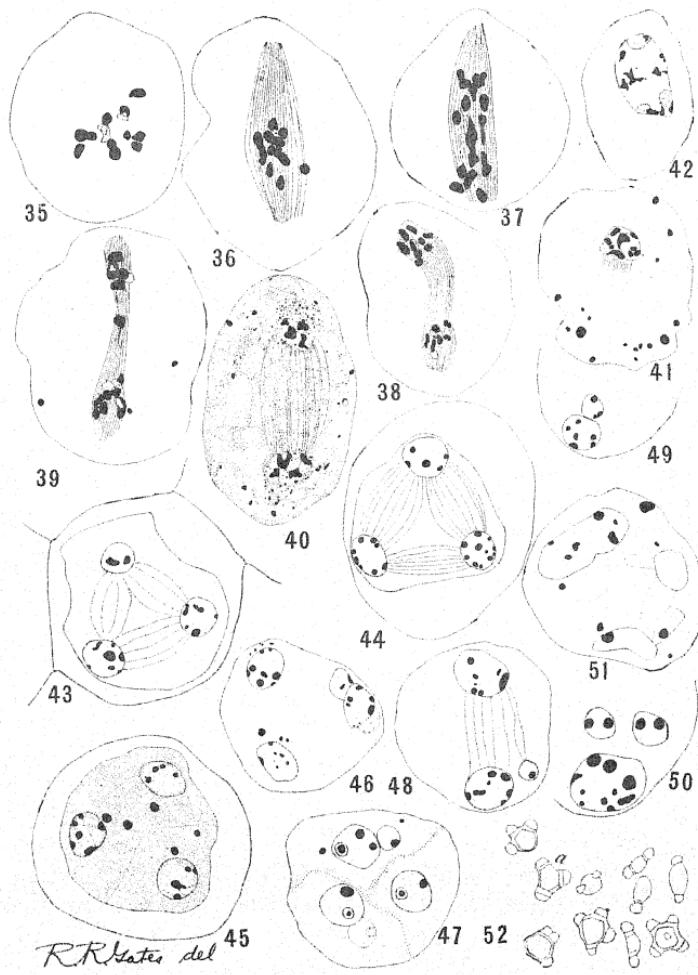




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## PLATE IV

FIGS. 35-42. *O. Lamarckiana* (dominant).

FIG. 35. Another equatorial view; the unequal appearance of the members of a pair may be due to foreshortening.  $\times 1500$ .

FIG. 36. Heterotypic spindle showing tendency of chromosomes to arrange themselves in groups of more than two.  $\times 1500$ .

FIG. 37. Early anaphase, showing viscid consistency of some of the chromosomes; the number is at least twenty; no heterochromosomes present; iron-haematoxylin-orange;  $7.5 \mu.$   $\times 1500$ .

FIG. 38. Late anaphase, showing premature splitting of the chromosomes for the homotypic division; twenty chromosomes present;  $7.5 \mu.$   $\times 1500$ .

FIG. 39. Late anaphase; one chromosome lagging behind on the spindle, and several chromatic bodies in the cytoplasm;  $7.5 \mu.$   $\times 1500$ .

FIG. 40. Telophase in which the reconstruction of the daughter nuclei has begun;  $7.5 \mu.$   $\times 1500$ .

FIG. 41. One of the daughter nuclei in the early stages of reconstruction; remnants of the spindle still connect it with the sister nucleus; several large chromatic bodies are found in the cytoplasm;  $7.5 \mu.$   $\times 1500$ .

FIG. 42. Polar view of daughter nucleus in late stage of reconstruction; iron-haematoxylin-orange;  $10 \mu.$   $\times 1500$ .

FIGS. 43-51. *O. lata* (dominant).

FIG. 43. Pollen tetrad, showing three of the four nuclei; the latter vary in size and in chromatin content; the tetrads are still attached to each other at the ends, but free laterally;  $5 \mu.$   $\times 1225$ .

FIG. 44. Same as last, but nuclei of nearly equal size; safranin-gentian-orange;  $5 \mu.$   $\times 1275$ .

FIG. 45. Tetrad showing first appearance of walls; chromatic bodies in the cytoplasm;  $5 \mu.$   $\times 1225$ .

FIG. 46. Tetrad showing two extra nuclei attached to one of the daughter nuclei;  $5 \mu.$   $\times 1275$ .

FIG. 47. Tetrad after wall formation, showing extra nuclei, and chromosomes in the cytoplasm not surrounded by a nuclear membrane;  $8 \mu.$   $\times 1225$ .

FIG. 48. Section of a tetrad, showing a small extra nucleus, formed by one chromosome, connected by spindle fibers with one of the larger nuclei;  $5 \mu.$   $\times 1225$ .

FIG. 49. Portion of a tetrad showing a small extra nucleus beside one of the tetrad nuclei;  $5 \mu.$   $\times 1225$ .

FIG. 50. Portion of tetrad showing two small extra nuclei, each containing two chromosomes;  $5 \mu.$   $\times 2125$ .

FIG. 51. Tetrad showing irregular nuclei, and chromatin in the cytoplasm;  $8 \mu.$   $\times 1500$ .

FIG. 52. Young pollen grains of *O. Lamarckiana* (dominant); *a* shows a normal-shaped grain; grains showing various abnormalities, such as those illustrated, are of frequent occurrence.  $\times 310$ .

DEVELOPMENT OF OVULE AND FEMALE GAMETO-  
PHYTE IN *GINKGO BILOBA*

CONTRIBUTIONS FROM THE HULL BOTANICAL LABORATORY  
XC

IDA ELEANOR CAROTHERS

(WITH PLATES V AND VI)

Ovules were collected at Elyria, Ohio, at intervals of two weeks, from the first of May until the latter part of August 1905, and were sent to Chicago packed in wet cotton. In April 1906, additional collections were made. Having been freshened by a stay in a moist chamber, the material was killed and fixed in chromacetic or chrom-acetosmic acid and put up in paraffin.

I wish to acknowledge my indebtedness to Miss A. M. STARR of Elyria, Ohio, for material, and to express my appreciation of the helpful suggestions of Professor JOHN M. COULTER and Dr. CHARLES J. CHAMBERLAIN of the University of Chicago, where the work was begun.

**Megasporangium**

DEVELOPMENT.—The megasporangia are borne on the short spur shoots, which bear also a few leaves, the longer shoots producing leaves only. By the first of April the terminal buds of the spur shoots have begun to swell slightly, and after removal of the brown bud-scales, the green leaves can be seen, but ovules are not yet distinguishable. Unopened buds collected the middle of April contain ovules which, with the stem bearing them, measure 2<sup>mm</sup> in length. The ovules alone are only 0.25<sup>mm</sup> long; they appear singly or in pairs at the end of leafless stems, and are pale cream color, while the leaves in the bud are bright green. Vertical sections (*fig. 1*) show the undifferentiated nucellus not yet wholly enclosed by the integument.

The buds soon open, and the leaves and ovule-bearing stems increase greatly in length. Not all the buds are fertile, but in those which are, the ovule-bearing stems vary in number from one to six. The ovules are still pale cream color, but show a faint green tinge which deepens rapidly as they enlarge. The ovule stalks appear, not at the growing tip of the spur shoot, but around it, as do the

strobili of most other gymnosperms. The succession of structures from without to the center of the bud is (*a*) brown bud-scales, (*b*) green scale-leaves, (*c*) foliage leaves, (*d*) ovules, (*e*) foliage leaves (*fig. 2*). Sections of these ovules show the nucellus enclosed by the integument but still entire, no tissue having broken down to form the pollen chamber.

At the beginning of May the ovules average 0.75<sup>mm</sup> in length and are little if any greater in diameter than the stems that bear them. In some cases the nucellar beak protrudes as a tiny papilla from the micropyle. Vertical sections (*figs. 3, 4*) show the presence of a large pollen chamber, often containing pollen grains already developing tubes.

**SPOROGENOUS TISSUE.**—In ovules collected the latter part of April and the first of May, the sporogenous tissue has been differentiated, and all stages from immature mother cells to complete tetrads are to be seen. About at a level with the junction of the nucellus and the integument (*figs. 3, 4*) is an ovoidal mass of cells (*fig. 5*) which differ from those around them in being larger, having granular cytoplasm without vacuoles, and large nuclei with conspicuous nucleoli. All these have the appearance of sporogenous cells, although as a rule there is only one mother cell (*fig. 5*). In fifty ovules only one shows more than a single mother cell (*fig. 6*), that single exception possessing two.

The origin of the mother cell from a hypodermal cell could not be traced. Nor could it be determined whether the differentiation of the mother cell precedes that of the surrounding cells, in which case they would be purely tapetal in morphology as well as in function; or whether the whole mass is differentiated, the picking out of the functional mother cell occurring later. If the latter be the case, the mass is sporogenous, and the presence of such cells around the youngest mother cell observed strengthens the view that such is the case. The origin of the two mother cells and the cells between them from the same row (*fig. 6*) is another point in favor of this probability.

The mother cell is usually slightly below the center of the mass (*fig. 5*), and is distinguishable from the surrounding cells by its greater size, more deeply staining wall, and non-granular cytoplasm. At first it is non-vacuolate and the large nucleus is near the center

(*fig. 7*), but later a large vacuole appears below and sometimes a smaller one above the nucleus, the latter consequently taking a position nearer the top of the cell (*fig. 8*). The mature mother cell shows, in the cytoplasm below the vacuole, a peculiar kinoplasmic mass (*fig. 8*) similar to those already reported in the mother cells of *Thuja* (7), *Taxodium* (1), *Larix* (6), and *Taxus* (2), and in the eggs of *Thuja* and *Taxodium*. The significance of this mass has not been determined.

The nucleus soon goes into synapsis (*fig. 9*), the kinoplasmic mass disappearing about the same time. In the single preparation showing the next stage, the eight chromosomes, which result from synapsis, divide even before the disappearance of the nuclear membrane and entrance of the spindle fibers, although the latter are visible in the surrounding cytoplasm (*fig. 10*). The chromosomes give no indication of their second division, although in some forms such indications are usual. Repeated counting of chromosomes on spindles in various stages through the prothallium gave the constant number eight.

**TETRAD.**—The spindle of the first division is peculiar in being obliquely placed (*figs. 11, 12*), possibly as a result of the vacuole in the lower part of the cell, the kinoplasmic fibers extending only in the cytoplasm next the wall. The lower of the two cells resulting from this division is slightly larger (*fig. 6*) than its sister. It soon markedly exceeds the other cell, however, the dividing wall arching up into the latter (*fig. 13*). The division of the lower cell (*figs. 14, 15*) precedes that of the upper, which in some cases does not divide at all (*fig. 14*). Usually, however, a linear tetrad is formed (*fig. 16*), the lowest spore of which enlarges rapidly, absorbing the contents of the other three. Although the linear arrangement of the four spores is most common, other forms of tetrads were observed. One case of a bilateral tetrad (*fig. 18*), and two instances of the transverse division of the lower cell followed by the vertical division of the upper cell were found. In one of the latter the nucleus of the upper cell has divided, but the separating wall has failed to appear (*fig. 9*). In all cases the lowest spore, the one nearest the chalaza, enlarges before the upper ones do, and by absorbing material from them and monopolizing most of the material coming into the ovule prevents their further development. Similar to this earlier development of the lowest spore of the tetrad is

the development of the lower of the two mother cells in a single ovule (fig. 6). COULTER (3) suggests that this early development of the lowest cell may be the result of a favorable position in regard to nutriment coming up through the chalazal tissue.

The functional spore enlarges rapidly, its nucleus being held near the upper end of the cell by a large vacuole below. The presence of this vacuole in the spore suggests the probability of the parietal position of the free nuclei of the embryo sac from the very beginning. The appearance of a vacuole and the consequent parietal placement of the two nuclei resulting from the first division of the spore nucleus is reported by LAND in *Ephedra trijurca* (8); but in Ginkgo the vacuole is present even before the formation of the first gametophytic spindle, being in fact the same vacuole that was in the mother cell.

**PROTHALLIUM.**—*Free nuclear division.*—Ovules of two weeks later are somewhat larger, and the embryo sacs contain free nuclei (fig. 20), the number ranging from 16 to 64. At this stage the sac is very thin, staining no more deeply than does an ordinary cell wall, and the plasmic layer lining it is very delicate. The nuclei are well separated and each has a conspicuous nucleolus. They are lenticular in side view and circular in face view, and average  $13.5 \mu$  in long diameter by  $8.14 \mu$  in short diameter. In these early stages the division of the free nuclei is simultaneous, although each of several sacs with 64 spindles shows the latter in several different phases (fig. 21). This is of interest, inasmuch as later free nuclear division proceeds irregularly, nuclei in a single sac showing all conditions through the various stages of karyokinesis and resting (fig. 24.) At first the nuclei of such sacs are definitely placed, so that although all stages are to be seen in the sac, the nuclei in any given portion are in the same stage. Later the sacs show nuclei in all stages grouped without regard to condition.

Free nuclear division continues from the second week in May until the first week in July, the whole ovule meanwhile enlarging and the embryo sac growing rapidly in both size and thickness. The protoplasm becomes granular and the free nuclei divide so rapidly that they decrease in size (figs. 22, 24). By the first of July the cytoplasm has become very granular (fig. 23), and the number of nuclei

large, the numerous divisions and their irregularity making the number larger than 256, which is so common a number among the gymnosperms. It is not impossible that the irregularity in division of the free nuclei, varying as it does from the simultaneous division said to obtain in other forms, may have been due to the unnatural conditions—growth after separation from the tree—although an effort was made to render conditions as natural as possible.

*Wall formation.*—About this time there appears on the outer surface of the plasmic sac a delicate membrane (*fig. 24*). This is not a *Hautschicht*, nor is it in any way attached to the original spore wall or embryo sac. It is a true cell wall of the type known as walls of deposit, and is formed by the protoplasm which lines it. Then walls appear in the cytoplasm, perpendicular to this enclosing membrane and with their outer edges fastened to it (*fig. 25*). The current accounts of wall formation following free nuclear division in the embryo sac, beginning with Miss SOKOLOWA's (10) description and continuing through subsequent papers by other investigators, state that the walls appear at right angles to the embryo sac or spore wall, and with their outer edges fastened to it. This is distinctly not the case in *Ginkgo*; and the ease with which the spore membrane may be peeled away from the prothallia of many other gymnosperms, even when the latter are but partly developed, suggests the probability that it is not the case in them.

It has long been known that the walls of microspores are quite separate and distinct from the wall of the mother cell in which they lie, and that the spores when mature escape from the mother cell membrane through ruptures or by its solution. In sectioned microsporangia showing developing tetrads, these separate surrounding walls of the mother cells may be readily seen. It is known also that the outer portion of the microspore wall, that next the wall of the mother cell, is formed by deposit, and not upon spindle fibers, as are the walls between the spores themselves. The megasporangium wall, as can be seen in the figures showing the tetrad formation, with the exception of that comparatively small portion which divides it from the non-functional spores, is the old membrane of the mother cell and corresponds to the mother cell wall enclosing the microspore tetrad. The formation of this separate membrane upon the outer surface of

the sac by deposit, therefore, corresponds to the formation of the walls of the microspore, and the fastening to it of the outer ends of the radial walls corresponds to the similar attachment of the walls cutting off the prothallial cells in the male gametophyte.<sup>1</sup>

These walls, at right angles to the enclosing membrane and fastened to it, separate the nuclei, probably each one being enclosed in a cell of its own. In other forms the first radial walls are said to come in on the spindles of the last simultaneous free nuclear division. Since the later divisions of the free nuclei here are far from simultaneous, this cannot be the case. Although the coming in of these first walls was not observed, the subsequent centripetal growth and division of the cells formed by their development are clearly seen (fig. 25). The inner cells of the centripetal rows are larger than the outer ones, the size of the latter being early diminished by repeated divisions, both tangential and radial (fig. 25). In most cases the nuclei of these cells, which are open to the interior of the sac (fig. 27), are much larger than those of the enclosed cells near the periphery. Most of the open cells have a single nucleus (fig. 27), but two nuclei are to be seen in some (fig. 28), and in a few, three nuclei. Binucleate cells (figs. 29, 30) are common in the tissue near the open central space, and even a few multinucleate cells are present (fig. 31), but since cells with a single nucleus are not found in the mature endosperm, these very cells must later become uninucleate. Whether this change results from a fusion of the several nuclei, a degeneration and absorption of the superfluous number, or their separation by the formation of intervening walls is not certainly known. In all cases of wall formation seen within cells, however, the wall is on a regular karyokinetic spindle. Usually the two nuclei in a cell are in contact, and it is not uncommon for one of them to be smaller and more dense than the other, suggesting its degeneration and absorption (fig. 30). From this it appears probable that cells which have at first two or more nuclei become uninucleate by a fusion of the several into one, or by the degeneration and absorption of the superfluous ones, and not by the formation of separating walls.

<sup>1</sup> One case of curious and irregular wall formation, abnormal and perhaps the result of pathological conditions, is seen in a sac of June 19. The plasmic layer containing free nuclei is as usual, but in addition there are eight little groups of nuclei, each group quite separate from the others and each with an inclosing membrane (fig. 26).

Wall formation in the cells open and growing toward the center, which is filled with sap, is carried on in the usual way at the peripheral end of each new cell, by the formation of a nuclear plate on the spindle of the division by which the new nucleus is formed (*fig. 32*).

The mode of formation of the side walls of these same cells, however, is not so easily determined. Curious double spindles (*fig. 33*) and extra fibers radiating from nuclei in the last stages of karyokinesis (*fig. 32*) indicate some such procedure as this: When a nucleus divides, lengthening the centripetal row, fibers radiate from it, not only to the sister nucleus, but radially as well, connecting with similar fibers radiating from other nuclei, and upon the resulting spindles are formed the plates which later develop into walls. This mode of wall formation has long been known in the endosperm of many angiosperms. In some cases are to be seen similar double spindles which lack nuclei at the two ends, the single nucleus concerned being in the middle. Further work is necessary to settle this point.

Centripetal growth continues through July and August, being most active at the base and filling the central cavity with tissue by the last week in August. In shape the prothallium is almost ellipsoidal, being slightly flattened on two opposite sides (*figs. 34, 35*). As a result of the shape and the equal growth from the sides, the closure of the tissue is in a plane parallel with the broader sides of the gametophyte. Only in sections cut at right angles to the flattened sides does the closure appear as a line (*fig. 34*). Upon approaching each other the open, centripetally growing cells of opposite sides do not unite and form a common end wall, as they are said to do in many other forms. Instead, each cell forms an independent end wall, separate from that of the other cells (*fig. 36*). The resulting body is not a solid mass, but a tissue which may be easily opened at the middle. Later these separate end walls, lying as they do against each other, may so unite as to appear and really form a common wall, but such is not the case in the oldest prothallia examined.

*Growth.*—Simultaneous with centripetal growth, the whole prothallium increases in bulk by growth and division of cells, the inner ones enlarging greatly and those at the periphery continuing meristematic (*figs. 25, 38*).

Archegonium initials appear very early; the two-celled neck has

been formed and the central cell has enlarged considerably before the tissue lining the sac equals in depth the width of the central cavity (*fig. 37*). HIRASÉ (4) has described the development of the archegonia.

The outer wall of the prothallium, the first wall built by the plasmic sac, becomes much thickened, and the megasporangium membrane or embryo sac becomes exceedingly thick and dense, making the entrance of the killing and the fixing fluids difficult. The structure and development of the megasporangium wall is set forth in a paper by THOMSON (11), but as my results differ slightly from his, I shall give them. THOMSON states the thickness of the megasporangium coat from a mature seed to be  $4.5-5 \mu$ . The wall shown in *fig. 22* measures  $2.1 \mu$  in thickness and that of an endosperm which has just closed in the center measures  $6.16 \mu$ . At first the membrane, which as shown above is in reality the mother cell wall, is thin and delicately granular (*fig. 20*). It rapidly thickens and becomes coarsely granular. A cross-section shows a very thin inner layer acting as a base for the transversely placed rods of the thick outer layer (*fig. 22*). THOMSON reports these rods as being quite irregularly placed. Subsequent thickening takes place in the outer layer only, the rods increasing in length and thickness, and the thin inner layer finally disappearing (*fig. 25*). Not infrequently, in material of this stage, little bunches of the rods forming the wall are found torn out and scattered over the slide. The result of the ease with which the outer layer is torn across between the rods and the resistance offered by the thin inner layer is shown at the upper end of the wall in *fig. 22*. The ends of the rods, presented at right angles at the surface of the spore coat, are the cause of the slight roughness there. After the disappearance of the smooth inner layer, the inner surface becomes similarly rough.

The complete independence of the heavy embryo sac and the outer wall of the prothallium explains the position of the former with reference to the archegonial chamber of the mature endosperm. Being entirely free from the tissue of the endosperm, it is lifted up by the growth of the tissue around the archegonial chamber until it is some distance from the floor of the latter and forms a roof over it.

From early in May the ovules are green throughout, but as they increase in size the inner tissue becomes hyaline, the chlorophyll being

abundant in one place only—a shallow region at the surface of the single heavy integument. Soon after the appearance of walls and long before the filling of the sac with tissue, the gametophyte becomes green. In a few weeks it is by far the greenest thing in the ovule. An alcholic solution of this green pigment gives the spectrum of chlorophyll. The presence of chlorophyll, evidently functional, within a gametophyte wholly enclosed within an ovule has not been reported before. The thin walls of the cells of the integument, the paucity of chlorophyll there, and the presence of numerous large cavities full of a clear viscid liquid favor the transmission of light to the gametophyte, and, as a meristematic tissue, it responds by the formation of the pigment. WARMING (12) reports that the endosperm of *Cycas circinalis*, if fertilization fails to take place, sometimes grows out through the micropyle and in the light becomes green. No mention, however, is made of its becoming green before protruding.

The cells of the endosperm early fill with starch, the large grains characteristic of the storage forms being abundant in the inner cells (figs. 26-38), while smaller grains in all stages of formation are to be seen in the periphery.

Some of this starch is undoubtedly manufactured by photosynthesis within the prothallium, but some may be the result of absorption from surrounding nucellar and integumentary tissues. Starch is plentiful, however, only in the outer layers of integument, never close to the endosperm.

#### Spongy tissue

The spongy tissue, in the midst of which the mother cell appears, performs an important function in the nutrition of the prothallium. As the megasporangium母 cell forms the tetrad, the whole spongy mass increases in bulk, the individual cells multiplying by spindles at right angles to the periphery of the mass (fig. 39). The nucellar tissue immediately surrounding at once shows signs of being absorbed, the protoplasmic contents of the cells becoming dark and granular even before the walls suffer collapse (fig. 39). This mode of increase in bulk of the spongy tissue and the adaptation of the surrounding nucellar tissue is described at length by Miss FERGUSON (4) in her work on *Pinus*. The result of this activity is a growing mass, at first

ovoidal, then ellipsoidal, of glandular cells encroaching upon and absorbing the surrounding nucellar tissue, the whole being enclosed in an enlarging cavity whose wall is made of elongating dividing cells. Although at first densely granular and non-vacuolate (figs. 5, 6), the absorbing cells soon become vacuolate (fig. 18), then multinucleate (fig. 22). Whether these nuclei are the result of direct or indirect division is not known. That the former is probably the case is suggested by the occurrence of such division in the absorbing cells of other sporangia. Moreover, spindles are seen in early stages only, and in those cases a plate is always present.

Until the fourth week in June the spongy tissue cells show great activity (fig. 22) in encroaching upon and absorbing the adjacent tissues, but soon afterward they themselves show signs of being absorbed by the enlarging prothallium (fig. 24), the latter being still in the free nuclear stage, but having formed the enclosing wall.

By the time the radial walls are formed and centripetal growth has brought the tissues one-third of the distance across the sac, the spongy tissue cells have been absorbed (fig. 25), their remains being only a thin mass of collapsed and heavily staining walls. The embryo sac now lies against the undifferentiated nucellar tissue, separated from the living cells of the latter only by this mass of dead cells which have given up their substance. When the prothallium has become a mass of tissue, it is quite near the surface of the nucellus, instead of far below it as at first. Most of the upper part of the nucellus has been absorbed, and the nucellar beak, at first so conspicuous, has collapsed (fig. 34).

#### Integument

The tissue of the single, thick integument is homogeneous at first, but by the last of May it is differentiated into three distinct tissues (fig. 40): an outer layer of large thin-walled cells with many mucilage-filled cavities; a middle layer of small, isodiametric cells; and an inner region of large, very thin-walled cells loosely held together. This inner layer is further differentiated into an outer layer having cells transversely elongated, and an inner layer having cells vertically elongated. This innermost layer of vertically elongated, delicate-walled cells appears only in the free portion of the integument, but the

remaining layers extend through its entire length, being the same both where it is separate from the nucellus and where it is continuous with it. The outer of these three layers becomes the fleshy, juicy part of the ripened fruit (*figs. 34, 35*); the middle layer, by a great thickening of walls and compacting of cells, becomes the stony coat; the inner fleshy layer, which is delicate and watery in early stages, becomes crushed and dry, forming the papery layer which lines the stony coat and adheres to it in the mature fruit. The nucellar tissue is quite distinct from the watery tissue of the integument, and in the upper part the two are not even in contact (*fig. 40*), so the former cannot form the papery coat as it is said to do by SEWARD and GOWAN (9). Below the line of junction of the nucellus and integument the former may contribute some small part to the papery coat, but it is almost entirely absorbed by the prothallium.

At the base of the ovule two bundles enter the watery tissue, coming up through a little gap in the tissue of a stony coat (*fig. 40*). Their course lies along the inner surface of the watery tissue, one curving up on each side and ending just below the point at which the integument becomes free from the nucellus. The abundant liquid in this watery tissue doubtless comes up through these bundles. Upon removal of the integument, which splits readily into two lips, the nucellus is seen to be flattened on two opposite sides, the intervening sides being angled (*figs. 34, 35*). It is in these angled sides that the bundles end (*fig. 35*). In some cases there is a three-lipped integument and a three-angled nucellus, there being also three bundles, one ending in each angled side.

#### SUMMARY

1. The ovule has a conspicuous nucellar beak and pollen chamber.
2. The sporogenous tissue is deep within the nucellus.
3. Differentiation of the mother cell has taken place by the first of May.
4. One mother cell is usual, but more may occur.
5. A peculiar kinoplasmic mass is present in the mother cell.
6. The gametophyte number of chromosomes is eight.
7. The first spindle is obliquely placed in the mother cell.
8. The tetrad may be complete or incomplete.

9. The tetrad is usually linear, but sometimes bilateral, or a combination of bilateral and linear.
10. The lowest spore is the functional cell.
11. The spore is vacuolate from the first and the free nuclei of following stages are probably always parietally placed.
12. Free nuclear division is at first simultaneous but gradually becomes irregular.
13. Free nuclear division extends, approximately, from the second week in May until the first week in July.
14. The cytoplasm, at first delicate, becomes granular and forms upon its outer surface a delicate wall.
15. Centripetally growing walls are formed between the nuclei and with their outer edges fastened to the membrane developed at the outer surface of the plasmic sac.
16. The sac fills with tissue by centripetal growth and division of cells, the inner ends being open.
17. These open cells are usually uninucleate but sometimes multinucleate.
18. Binucleate and multinucleate cells are frequent in young prothallial tissue, but later they become uninucleate, probably by fusion of the several nuclei or by degeneration of the superfluous ones.
19. When the centripetally growing cells meet at the center each forms an independent end wall.
20. The megasporangium wall and the outer wall of the prothallium become much thickened.
21. The megasporangium wall is composed of an inner, thin, firm layer, and a very thick, outer layer made up of rods formed at right angles to the surface.
22. The archegonia are quite far developed while there is still a large central cavity.
23. The gametophyte develops abundant chlorophyll, becoming the greenest tissue of the ovule.
24. The spongy tissue surrounding the mother cell is tapetal in function, absorbing the surrounding tissue, and finally being itself absorbed by the growing prothallium.
25. The spongy tissue cells become vacuolate and multinucleate.
26. The integument is early differentiated into three distinct

tissues, which become respectively the fleshy coat, the stony coat, and the papery coat.

27. Abundant liquid is brought to the watery layer by two bundles which enter through a gap in the stony coat and terminate just below the point of separation of the integument and nucellus.

CHARLESTON, ILL.

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#### EXPLANATION OF PLATES V AND VI

With the exception of figs. 3, 4, 34, 35, 37, and 40, the figures were drawn with the aid of an Abbé camera lucida and all were reduced one-half in reproduction.

#### PLATE V

FIG. 1. Vertical section of a single ovule; April 15, 1906.  $\times 15$ .

FIG. 2. Habit sketch of spur shoot, showing opened fertile bud; April 25, 1906.  $\times 2$ .

FIG. 3. Vertical section of a pair of ovules; May 1, 1905.  $\times 12$ .

FIG. 4. Vertical section of a single ovule; May 1, 1905.  $\times 12$ .

FIG. 5. Vertical section through sporogenous mass with single mother cell; May 1, 1905.  $\times 325$ .

FIG. 6. Vertical section through sporogenous mass with two mother cells; May 1, 1905.  $\times 325$ .

FIG. 7. Young mother cell; May 1, 1905.  $\times 650$ .

FIG. 8. Mature mother cell, showing vacuoles and kinoplasmic mass; May 1, 1905.  $\times 650$ .

FIG. 9. Mother cell in synapsis; May 1, 1905.  $\times 650$ .

FIG. 10. Mother cell just after synapsis, showing the eight chromosomes already divided for the heterotypic division, and the spindle fibers in the cytoplasm; May 1, 1905.  $\times 650$ .

FIG. 11. Mother cell showing oblique spindle of heterotypic division; May 1, 1905.  $\times 650$ .

FIG. 12. Mother cell showing oblique division; May 1, 1905.  $\times 650$ .

FIG. 13. First division of mother cell completed; May 1, 1905.  $\times 650$ .

FIG. 14. Lower cell showing spindle of homotypic division; upper cell preparing for division; May 1, 1905.  $\times 650$ .

FIG. 15. Division of lower cell completed; May 1, 1905.  $\times 650$ .

FIG. 16. Incomplete tetrad; upper cell failing to divide; May 1, 1905.  $\times 650$ .

FIG. 17. Linear tetrad; lowest cell enlarging; May 1, 1905.  $\times 650$ .

FIG. 18. Bilateral tetrad; May 1, 1905.  $\times 650$ .

FIG. 19. Combination linear and bilateral tetrad; wall failing to appear in upper cell after vertical division of nucleus; May 1, 1905.  $\times 650$ .

FIG. 20. Vertical section of a sixteen-nucleate sac and tapetum; May 18, 1905.  $\times 225$ .

*PLATE VI*

FIG. 21. Portion of sac showing spindles of free nuclear division in slightly different phases; May 18, 1905.  $\times 650$ .

FIG. 22. Portion of embryo sac showing granular cytoplasm, free nuclei, and multinucleate tapetal cells encroaching upon nucellus; June 5, 1905.  $\times 650$ .

FIG. 23. Face view of plasmic sac with free nuclei; June 5, 1905.  $\times 650$ .

FIG. 24. Portion of embryo sac showing absorption of tapetum; free nuclei in different conditions; and a cell wall developed upon the outer surface of the plasmic sac; June 20, 1905.  $\times 650$ .

FIG. 25. Section of prothallium, showing centripetal walls fastened to outer membrane which is separate and distinct from embryo sac; July 19, 1905.  $\times 325$ .

FIG. 26. Peculiar case of wall formation; June 5, 1905.  $\times 650$ .

FIG. 27. Open cell from inner end of centripetally growing row; July 19, 1905.  $\times 325$ .

FIG. 28. Open inner cell with two nuclei; July 28, 1905.  $\times 325$ .

FIG. 29. Binucleate cell from prothallial tissue; July 28, 1905.  $\times 325$ .

FIG. 30. Binucleate cell from gametophyte; one nucleus apparently undergoing absorption; July 19, 1905.  $\times 325$ .

FIG. 31. Multinucleate cell from prothallial tissue; July 28, 1905.  $\times 325$ .

FIG. 32. Open inner cell showing centripetal growth by means of spindles; extra fibers being attached to outer nucleus; July 19, 1905.  $\times 325$ .

FIG. 33. Curious incomplete double spindle from open inner cell; July 19, 1905.  $\times 325$ .

FIG. 34. Vertical section of ovule, showing embryo sac filled with tissue; the closure appearing as a line; August 21, 1905.  $\times 1$ .

FIG. 35. Transverse section of ovule, showing embryo sac filled with tissue; showing radiation of cell rows from line of closure; August 21, 1905.  $\times 1$ .

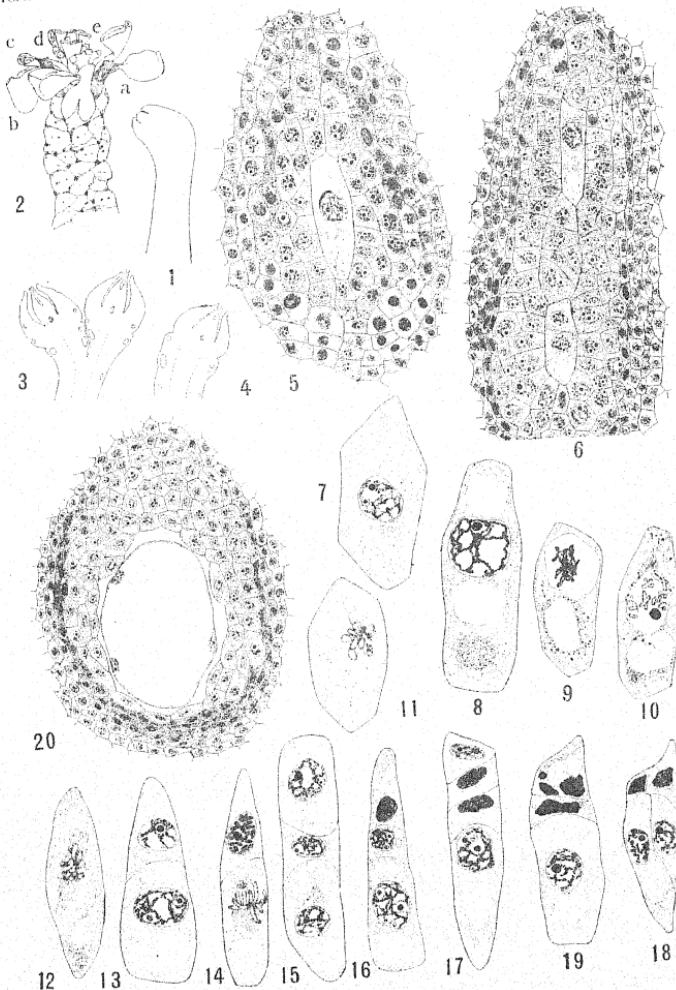
FIG. 36. Vertical section of tissue at line of closure, showing independent end walls of opposite cells; August 21, 1905.  $\times 325$ .

FIG. 37. Diagram of vertical section of prothallium, showing amount of tissue present at time when archegonia are becoming conspicuous; July 19, 1905.  $\times 2$ .

FIG. 38. Typical mature cell from gametophyte, showing single nucleus and abundance of starch; August 21, 1905.  $\times 325$ .

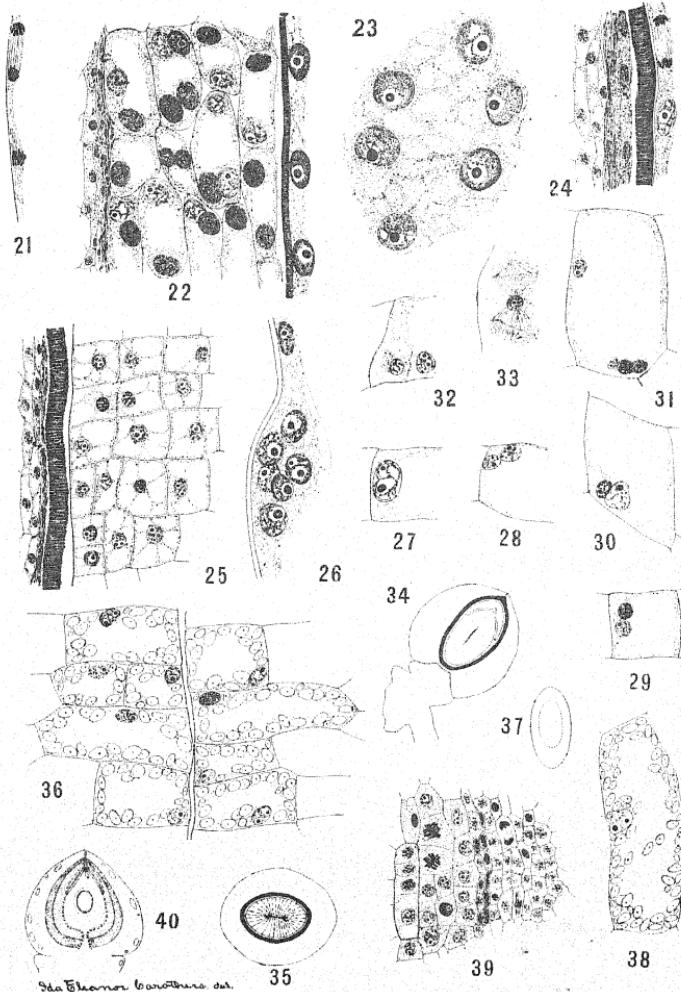
FIG. 39. Vertical section through spongy mass, showing mode of increase in size, and effect upon surrounding cells; May 1, 1905.  $\times 325$ .

FIG. 40. Diagram of vertical section of ovule, showing differentiation of tissues in the integument; June 10, 1905.  $\times 4$ .

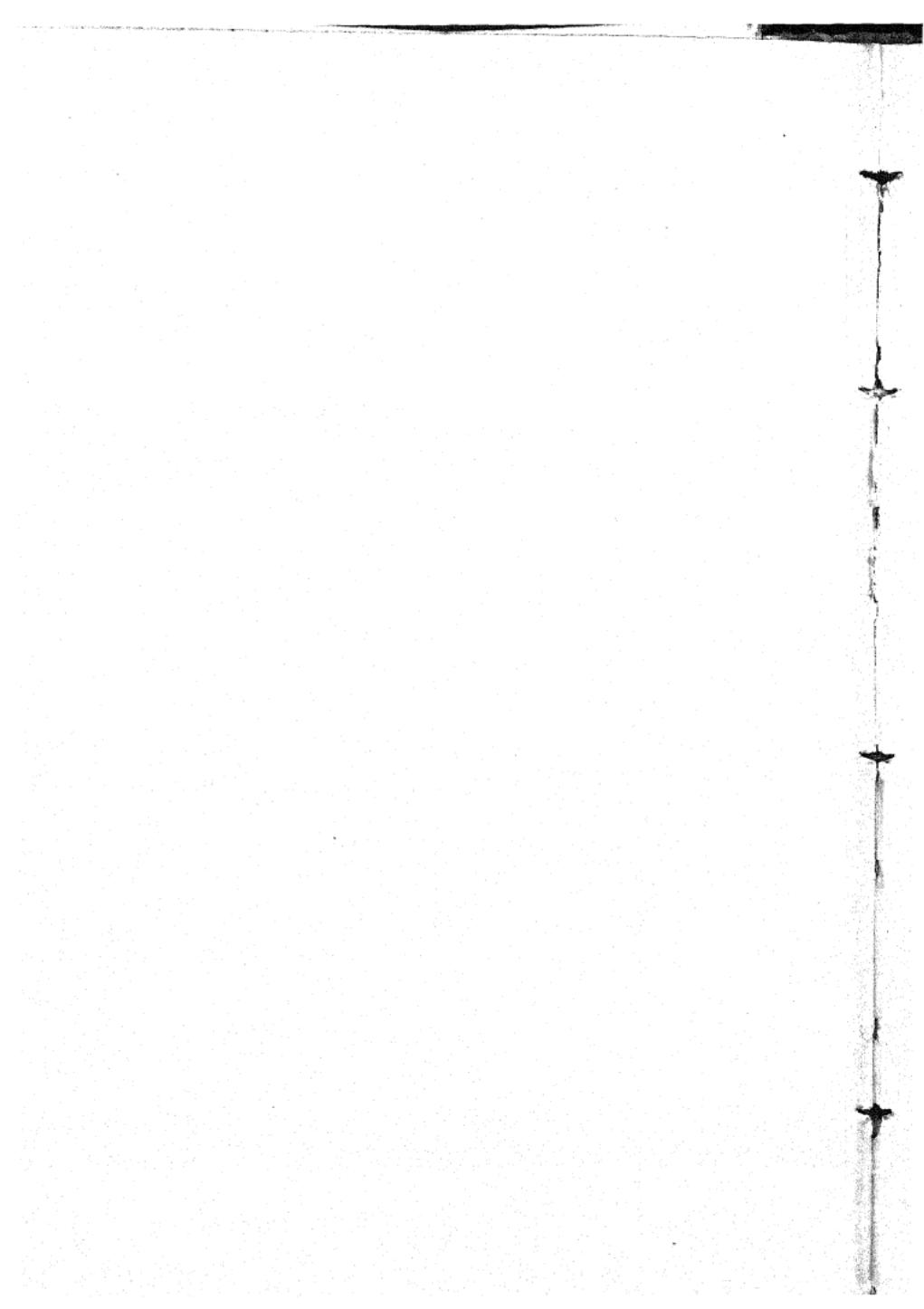


GAMETOPHYTES on GINKGO





CAROTHERS on GINKGO



## MORPHOLOGY OF SPATHYEMA FOETIDA

JAMES ELLIS GOW

(WITH SEVEN FIGURES)

During the late winter and the spring of 1906 the writer collected, in the vicinity of Blairstown, N. J., the material on which the following observations are based. The anthers of Spathyema and the portion of the receptacle in which the ovary lay imbedded were cut out, killed, and fixed in picracetic,<sup>1</sup> chromacetic, or picric acid, imbedded, sectioned, and mounted in the usual manner. The stains used were safranin, Delafield's haematoxylin, and Haidenhain's iron-alum haematoxylin. Of these the last gave by far the best results.

During the summer the buds which give rise to the plants of the next year develop on the thick underground rootstocks. By November the shoots are several inches in length. The flower buds occur as lateral offshoots of the leaf bud, in the outer integuments of which they are enclosed. Only one flower makes its appearance in connection with each shoot. Occasionally this one is lacking, and the leafy shoot appears alone.

### THE MICROSPORANGIUM.

During February growth is resumed in the flower bud. At this time the single anatropous ovule makes its first appearance in the form of a group of cells projecting obliquely downward into the cavity of the ovary. The stamens appear as short projections from the surface of the receptacle. The filaments do not begin to lengthen until considerably later. The anthers at this stage of development have not attained their full size, but the four microsporangia and the connective may be readily distinguished by the naked eye. Upon making a microscopic examination of the anther in cross section it is seen that the various portions are already well differentiated. Through the middle of the rather thin-walled cells forming the connective

<sup>1</sup> A saturated solution of picric acid to which is added 1 per cent. of glacial acetic acid.

runs a well defined vascular bundle. The whole anther is invested by a layer of epidermal cells whose outer surface is somewhat thickened. In the interior of each microsporangium is a mass of thin-walled sporogenous cells (usually fifteen to twenty), indicating that the archesporial cells have developed some time before. These are surrounded by a well-defined tapetal layer. The four microsporangia are borne on the ventral surface of the stamen.

During the latter part of February and the first week in March the anthers increase greatly in diameter. This increase does not take place in the length of the connective so much as in the size of the four microsporangia. The connective appears to have reached its full development earlier. The growth of the microsporangia is accompanied by rapid increase in the number of sporogenous cells. While in a resting condition, the sporogenous cells differ from the surrounding cells in their greater size, in their generally hexagonal shape, and in the fact that the nucleus is larger in proportion to the size of the cell and shows a more distinct chromatin network than does the nucleus of the vegetative cell. Division of the sporogenous cells continues until they form, taken together, two-thirds of the diameter of the microsporangium. The last cells formed are considerably larger than those formed in the earlier stages of the process.

While this is going on, the filaments begin to lengthen out and push the anthers up until they press against the infolded edges of the perianth. By the second or third week in March the divisions of the sporogenous cells are completed. The mature pollen mother cells thus formed are from one and a half to two times the size of the cells from which they have descended. The tetrad formation is as usual. The walls of the spores are greatly thickened and do not stain readily with any of the anilin stains.<sup>2</sup> The cytoplasm is granular and the nucleus ill-defined. From the difficulty experienced in finding material in this stage of development, it is evident that it is very evanescent.

An examination of the pollen grains in water shows that the exine

<sup>2</sup> Staining the tetrads with haematoxylin was not attempted. Sections fixed in picric acid and stained with eosin showed the fibrous portion of the anther stained bright orange, mature pollen grains deep red, tetrads light yellow.

is of appreciable thickness (though much thinner than the wall of the cell in which it was developed) and is covered with minute spiny projections. The vegetative and generative nuclei appear faintly when the interior of the grain is brought into focus. When the pollen grain is treated with a solution of iodine green they appear much more clearly, and it is seen that the latter is much the larger of the two. The division between the two takes place just before the expulsion of the pollen grain from its investing cell.

After the expulsion of the pollen grains the collapsed mother cells may be seen lying about and partially filling the cavity of the anther; but in a day or two they break down and disappear. Simultaneously with the disappearance of the mother-cell walls, the tapetal layer breaks down and the contents of the cells become scattered throughout the cavity of the microsporangium. By the beginning of the last week in March the microsporangium is filled with a mass of mature pollen grains. In the meantime the filaments, as a rule, have lengthened sufficiently to push the anthers out past the infolded edges of the perianth. When this has taken place the anthers dehisce and the pollen grains are set free. Frequently, however, the lengthening of the filament is delayed and the mature pollen grains remain in the anthers for a week or more before they are finally set free. This is a phase of the subject that must be reserved for future study, but it would seem that the development of the filament and dehiscence of the anther are dependent to a certain extent on weather conditions. Probably, too, there is a correlation between their development and the development of the pistil. The dehiscence is along two longitudinal creases, each one of which separates the two microsporangia lying at the same end of the connective.

#### THE MEGASPORANGIUM

Material gathered early in February shows the ovary as a tiny cavity in the tissue of the receptacle, beneath the short thick style. The ovule at this period appears as a group of thin-walled, undifferentiated, strongly nucleate cells, projecting downward into the ovary and nearly filling it. As yet there is no distinction of nucellus and integuments. The style has pushed up far enough to bring the partially developed stigmatic cells even with the edges of the sepals.

By the last week in February the cavity of the ovary has considerably increased and made room for the growing ovule. At this point the differentiation between the nucellus and its integuments begins to be noticeable, but as yet there is no division between inner and outer integuments. By the first or second week in March the outer integument begins to appear, and the inner integument grows up so as nearly to cover the nucellus, leaving only a narrow micropyle above its tip. An examination of the nucellus at this stage shows that it is invested by a distinct outer layer of cells differing from the interior cells in their greater regularity of outline. The cell at the tip of the

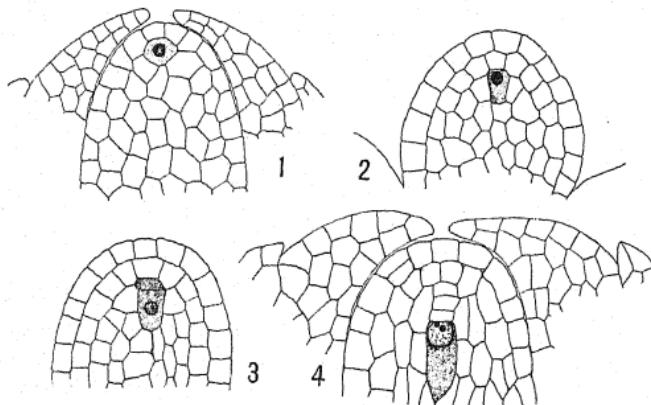


FIG. 1.—Ovule, showing archesporial cell. FIG. 2.—Same, after formation of two epidermal layers. FIG. 3.—First division of archesporial cell. FIG. 4.—The row of megasporangia. All  $\times 350$ .

nucellus and immediately under this investing layer is the archesporium (*fig. 1*). At this stage it differs from the surrounding cells neither in size nor in the nature of its contents, and can only be distinguished by its position.

As the nucellus grows, the cells of the outer layer divide, forming two layers outside the archesporium (*fig. 2*). The latter then divides transversely, cutting off a narrow tapetal cell (*fig. 3*). Up to this point the archesporial cell has undergone very little increase in size, but now the primary sporogenous cell begins to lengthen down-

ward rapidly, crowding the cells in the base of the nucellus, some of which break down. At the same time it undergoes two more transverse divisions, forming a row of three megasporocytes, the innermost of which functions (fig. 4).

The reduction division and the successive divisions up to the fertilization stage of the embryo sac proceed as usual (fig. 5-7). In the reduction division eight chromosomes were observed, and sixteen were counted in the vegetative cells. During the maturing of the embryo sac, it lengthens and becomes from one and a half to two times as

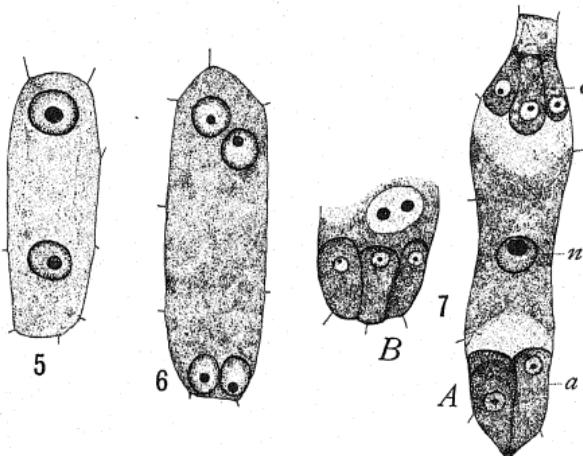


FIG. 5.—First nuclear division of embryo sac. FIG. 6.—Second nuclear division of embryo sac. FIG. 7.—A, mature embryo sac; *o*, oosphere; *n*, primary endosperm nucleus; *a*, antipodal cells; *B*, antipodal cells, and endosperm nucleus with two nucleoli. All  $\times 900$ .

long as it was at the first division of the megasporocyte nucleus. The increase is all toward the chalaza, with very little lateral increase. In fact, as the sac elongates it frequently contracts in the middle.

Of the material examined early in March, very few ovules were found in which the division of the archesporial cell had taken place. In many of the ovaries examined about April first the embryo sac was in the fertilization stage.<sup>3</sup>

<sup>3</sup> I give the earliest date at which the plant develops to the stage of growth described. As a matter of fact, many plants are belated, and flower buds may sometimes be found in the middle of April still unopened; but this is exceptional.

## FERTILIZATION

The process of fertilization takes place during April or early in May. It may follow quickly upon the completion of the division of the nuclei in the embryo sac, or the latter may remain for several weeks before being fertilized. All depends upon the accident of pollination. *Spathyema* is fertilized by the agency of insects, especially certain spiders which make a practice of spinning their webs inside the spathe and flies which are attracted by the rank odor of the plant.

The exact length of time required for the penetration of the pollen tube into the embryo sac was not determined. The stigmatic surface consists of a mass of loosely compacted cylindrical cells, thin-walled and not distinctly nucleated. Four vascular bundles run through the thick pyramidal style, corresponding to its four angles. The central portion of the style consists of a loose mass of thin-walled cells, through which the pollen tube readily forces its way to the upper end of the ovary. It then follows either the inner surface of the ovary or the outer surface of the integuments of the ovule until opposite the micropyle, through which it passes.

## THE EMBRYO

After fertilization, the oospore by transverse divisions forms a proembryo of four cells. The terminal cell then divides longitudinally, followed by a second longitudinal division at right angles to the first. Each of the four terminal cells then divides transversely. The most advanced material examined during the second week in May showed the embryo in this stage of division. Beyond that point it has not as yet been possible to carry the investigation.

BLAIRSTOWN, N. J.

## BRIEFER ARTICLES

### PRELIMINARY NOTE ON CERATOZAMIA

With the aid of a grant made by the Botanical Society of America at the New Orleans meeting in December 1906, the writer has been able to secure material for a study of Ceratozamia in most phases of its life history. In September 1906 a trip was made to the barrancas of the Almolonga Valley near Xalapa, Mexico, where the plant is quite abundant. Photographs and field notes were secured and material was fixed for a detailed morphological and cytological study. Arrangements were also made for obtaining material at frequent intervals.

In habitat Ceratozamia differs decidedly from Dioon, for Dioon is in the open, exposed to blazing sunlight, while Ceratozamia is in densely shaded places. The difference in light will be appreciated from the fact that a photographic plate which would be well exposed for Dioon in one-fifth of a second would require three minutes exposure for Ceratozamia. On the whole, Dioon and the plants associated with it are xerophytic; while Ceratozamia, though not found in wet situations, is associated with a luxuriant vegetation.

Stamineate cones which arrived in Chicago on March 10, 1906, shed their pollen within a week. Fertilization takes place more than a year after pollination. The first motile spermatozoids were observed June 9, 1906, and in the last week of June nearly every nucellus showed one or more pollen tubes in which spermatozoids were swimming. The spermatozoids were observed with diminishing frequency during the first two weeks in July. Swimming spermatozoids had previously been observed in Cycas by IKENO and in Zamia by WEBBER. Our observations add Dioon and Ceratozamia to the list. A section by LANG proves that the spermatozoid of Stangeria is also motile. Nothing is known of the pollen tube structures of the other four genera of cycads.

The ovulate cones disintegrate and free the seeds soon after fertilization, while the embryo is still of the same diameter as the filamentous suspensor. The seed has no resting period, but growth is continuous from fertilization to the leafy plant.—CHARLES J. CHAMBERLAIN, *The University of Chicago*.

## CURRENT LITERATURE

### BOOK REVIEWS

#### Progress of botany

In connection with the Vienna Congress, a series of papers describing recent progress in the various fields of botanical activity is being published under the editorship of Dr. J. P. Lotsy. The general title of the series is *Progressus rei botanicae*, and the first part, comprising four papers, has just appeared.<sup>1</sup>

ONTOGENY OF THE CELL.—Under this title STRASBURGER<sup>2</sup> gives an interesting and valuable history of the progress of cell studies since 1875. The various problems which in their turn have been most prominent in cytological investigation are treated with a breadth and grasp which would be impossible except for one who has contributed to the subject during its entire development. Particularly interesting is the treatment of problems like the centrosome, the origin of the spindle, double fertilization, etc., which have passed through the stage of dispute and polemic to comparative agreement. The treatment of the centrosome, which just now is perhaps the most investigated feature of the cell, is suggestive, the earlier problems, which are practically settled, being clearly marked off from the seemingly more difficult problems which are now occupying the attention of investigators. Text cuts are numerous and the bibliography is quite complete.—CHARLES J. CHAMBERLAIN.

PRESENT POSITION OF PALEOZOIC BOTANY.—SCOTT<sup>3</sup> has contributed a most timely account of the present status of our knowledge of paleozoic plants. The recent progress in this field has been most remarkable, and Professor SCOTT's connection with it has been conspicuous. Naturally the chief attention is given to the vascular plants, JEFFREY'S division of them into Lycopida and Pteropsida being used for convenience of presentation. Under the first division, Sphenophyllales, Equisetales, Psilotales, and Lycopodiales are presented; and under the second, Filicales, "Pteridospermeae," and "Gymnospermeae." Chief interest centers about the Filicales and pteridosperms. The question as to the existence of paleozoic ferns is answered decidedly in the affirmative; but with the statement that at present our knowledge of the group centers in the Botyopteridæ, "which have now come to be of supreme interest in the geological history of vascular plants." The development of our knowledge of pteridosperms is

<sup>1</sup> *Progressus rei botanicae*. Herausgegeben von der Association Internationale des Botanistes. Redigiert von Dr. J. P. Lotsy. Erster Band. Erstes Heft. pp. 317. Jena: Gustav Fischer. 1907. M. 18.

<sup>2</sup> STRASBURGER, EDUARD, Die Ontogenie der Zelle seit 1875. Op. cit. 1:1-138.

<sup>3</sup> SCOTT, D. H., The present position of paleozoic botany. Op. cit. 1:139-217.

told briefly and clearly, and their separation from gymnosperms as a coordinate group is defended, the provisional diagnosis being as follows: "Male and female sporophylls little differentiated from the vegetative foliage; no cones formed. Anatomy of either stem, or leaf, or both, of a Filicinean type, as was also the habit." It is hard to see that such characters are more important than those which distinguish the acknowledged groups of gymnosperms, and of equal importance with those that distinguish gymnosperms from angiosperms. The relations of the pteridosperms to the Cycadophyta and to the Cordaitales are also discussed. The whole paper is an admirable résumé of our knowledge of the subject, and will serve to present it clearly to many botanists who have either no time to consult the numerous original papers or no access to them.

BIBLIOGRAPHY OF PALEOZOIC FOSSIL PLANTS.—The third paper is an exceedingly useful bibliography prepared by ARBER.<sup>4</sup> There is first a list of general monographs and textbooks, followed by a list of memoirs on special subjects, after which the titles are arranged stratigraphically, beginning with the Silurian. Under each geological period, beginning with the Devonian, the arrangement is by plant groups and by countries.

GEOGRAPHIC BOTANY.—The fourth paper is by FLAHAULT.<sup>5</sup> After a general historical and explanatory introduction, the subject is presented under the following captions: (1) Descriptive phytogeography (Floristics), (2) Physiological phytogeography (Ecology), (3) Ontogenetic phytogeography, and (4) Historical phytogeography. The second topic occupies the largest space, the principal subtopics being discussions of the ecological factors, temperature, water, light, soil, humus, and the various so-called climates. Ontogenetic phytogeography deals with the history of the distribution of plants, including the historical origin of groups, migrations, and the succession of floras. Under historical phytogeography the work of man in relation to floras is discussed, including such subjects as the sources of rivers, mountain forests, dunes, etc. The paper brings together in an organized form the study of vegetation from the various points of view that have been often lumped under the general and very indefinite title Ecology.

—J. M. C.

#### A South African textbook in botany

This is not the first elementary textbook written for use in the South African schools, but it is perhaps the first to give so much attention to the ecological point of view.<sup>6</sup> In this respect it is to be compared with the elementary text-

<sup>4</sup> ARBER, E. A., NEWELL, Bibliography of literature on palaeozoic fossil plants, including some of the more important memoirs published between 1870-1905. Op. cit. 1:218-242.

<sup>5</sup> FLAHAULT, CH., *Les progrès de la géographie botanique depuis 1884, son état actuel, ses problèmes*. Op. cit. 1:243-317.

<sup>6</sup> STONEMAN, BERTHA, *Plants and their ways in South Africa*. 8vo. pp. ix+283. London: Longmans, Green, & Co. 1906. \$1.10.

books of COULTER or BERGEN. It is refreshing to miss the old familiar cuts that have done service for so long. Not only do the photographic reproductions represent South African scenes, but even the diagrams and cuts are made from South African plants, and for the most part especially for this book. The first chapters deal with seeds and germination, growth and duration of the vegetative organs, and various fundamental physiological topics. The chapter on plant defenses is illustrated by some of the remarkable desert xerophytes of the region, such as Crassula and Mesembryanthemum. Then follow chapters on vegetative reproduction, climbing plants, and migration. After a rather full consideration of flowers and fruits, there is a simple but useful key to the more conspicuous plants of the region. The old-fashioned order of BENTHAM and HOOKER is used in the classification, but this is probably justified by the fact that South African systematic works have followed the BENTHAM and HOOKER scheme. The book should do its part in making botany in the schools a more living and attractive subject.—HENRY C. COWLES.

#### MINOR NOTICES

**Spontaneous heating.**—The “heating” of hay and compost heaps has been an interesting biological phenomenon as to whose causes many were content to reason, but few to experiment. MIEHE recently published a scientific summary of his work, which is now followed by a booklet, addressed to a wider audience.<sup>7</sup> It embodies his own investigations for the past two years, to which are added the facts already known, so as to make a complete discussion of this subject, which is of both biological and practical interest.

The topics treated are: The course and maximum of the rise of temperature; chemical changes in the heating of compacted hay with exclusion of oxygen; cause of heating; culture substrata and researches with pure cultures; description of the nine most important organisms found in hay; self-sterilization by heating; the conditions of existence of thermophilous organisms in nature; heating plant materials as brood-beds for pathogenic organisms; relation of the fermentation of tobacco to the heating of hay; respiration and warmth; heating and spontaneous combustion, present and past.

The book will be particularly useful to men in experiment stations who are concerned with this problem, as it forms an excellent summary of present knowledge. It is handicapped by lacking an index, though it has an exhaustive bibliography.—C. R. B.

**The origin of species and varieties by mutation.**—It is an encouraging sign when such a scientific treatise as Professor DEVRIES' *Species and varieties* comes to a second edition within one year.<sup>8</sup> Probably no work on evolution written in

<sup>7</sup> MIEHE, HUGO, Die Selbsterhitzung des Heus. 8vo. vi+127. figs. II. Jena: G. Fischer. M3.50.

<sup>8</sup> DEVRIES, HUGO, *Species and varieties; their origin by mutation*. Edited by D. T. MACDOUGAL. Second edition. 8vo., pp. xviii+847. Chicago: The Open Court Publishing Co. 1906.

English has had such a great sale, at least for many years. Very few changes are to be seen in the new edition. The frontispiece shows Professor DEVRIES at the Desert Laboratory in somewhat informal garb. The few errors of the first edition have been corrected, and some alterations have been made for the sake of clearness. The most important new feature is an explanatory note on variations in *Oenothera biennis*. It is to be hoped that the sale of this second edition will also be great enough to necessitate the publication of yet another edition. In this way it will be possible for the general public to keep conversant with the rapid advance of our knowledge concerning mutation.—H. C. COWLES.

#### NOTES FOR STUDENTS

**Fixation of atmospheric nitrogen.**—In agricultural practice this is a most important matter; witness the many efforts to secure it in leguminous crops by inoculating the soil. Two recent elaborate papers concern themselves with the organisms which are supposed to do this in the soil, and from these studies it is clear that our knowledge of this process and its conditions rests upon most insecure foundations.

WARMBOLD, reviewing the literature relating to the alteration of the N-content of uncultivated soils, finds such a contradiction between the data, particularly those of BERTHELOT, who found N-fixation active in summer but nil in winter, and KOCH, who found it notable in winter, that he endeavored to determine the conditions for the process in nature, especially in relation to temperature, water content of soil, aeration (dependent on porosity of soil), and the amount and character of organic substances present.<sup>9</sup> Having carefully considered the sources of error in the methods of determining N in the soil, he believes his analytic results may be relied on; and with a warning against too wide generalization from his data he concludes: (1) Well-aerated soils may be enriched in N without the intermediation of organisms, this having occurred in two series of experiments, while in three others under apparently like conditions it did not take place. (2) Temperature exercises no obvious influence, nor does water content, provided the soil is in thin layers and well aerated. When, however, it is in masses and contains less than 20 per cent. water, accumulation of N is either small or a minus quantity; at 10 per cent. there may be marked loss of N. This difference in behavior depends on a difference in the N-compound in question. With 3 per cent. water or less, the loss in N is due not to denitrification by organisms, but is purely chemical. In sterilized soil 3-20 per cent. of water has no effect; at 30 per cent. N diminishes.<sup>10</sup> (3) Aeration had no discernible effect in small masses of soil, but it was advantageous in larger quantities containing 15 per cent.

<sup>9</sup> WARMBOLD, H., Untersuchungen über die Biologie stickstoffbindender Bakterien. Ein Beitrag zur Kenntnis der Veränderungen in Stickstoffgehalte des unbewohnten Ackerbodens. Landw. Jahrb. 35:1-123. 1906.

<sup>10</sup> But cf. as to this point SCHULZE on the effects of sterilizing soils. Abstract in BOT. GAZETTE 42:502. 1906.

water. (4) Artificially prepared humus neither affected the N-fixation favorably, nor could the bacteria use it as a source of food. (5) The N-fixing power of supplies of Azotobacter derived from pure cultures and grown in identical conditions was extraordinarily different at optimal temperatures as well as at low and higher points. Some light may be cast on this by the following.

In an elaborate examination of the behavior of Azotobacter<sup>11</sup> THIELE believes that he has established incontestably that this organism is capable of accumulating N in the laboratory; but he is quite uncertain whether this power belongs specifically to it, as for example alcohol production does to yeast. It is not impossible that N-starvation in the artificial culture or the stimulation by abundance of organic matter awakens an inherent capacity of Azotobacter to fix N, which ordinarily slumbers. The growth of Azotobacter in artificial cultures is neither decisive nor typical. Its mode of action in the soil is still entirely unknown and is likely to remain so, in spite of theories, until there have been devised more exact methods of investigating the extremely minute variations of the N in soils. In view of all this uncertainty, THIELE deprecates giving agriculturists any advice which would lead them to attempt to replace Chili saltpeter by a bacterial "fertilizer."—C. R. B.

**Burbank's work.**—An interesting and illuminating account of the breeding experiments of LUTHER BURBANK is contributed by DEVRIES to the *Biologisches Centralblatt*.<sup>12</sup> It is the first statement we have seen addressed to scientific men by a man competent to appreciate both the practical and scientific aspects of BURBANK's work.—C. R. B.

**Anti-enzyme.**—With the aid of BERTEL, CZAPEK<sup>13</sup> has now elaborated the results of ten years of research devoted to developing a chemical test for tropistic sensation. Until 1897, when the author published his initial paper of this investigation, we had no way of knowing that an organ had perceived a stimulus unless it manifested response in the form of a motor reaction. In fact it was assumed that perception had not occurred unless such a motor reaction followed. FITTING, in his work on tendrils, was able by indirect methods to show that an organ may perceive a stimulus and still be incapable of executing a motor reaction. In animal irritability it has long been believed that sensations involve alterations in the metabolism of the organ. To CZAPEK belongs the honor of a fruitful pioneer research. Root tips contain tyrosin, an unstable derivative of proteids, continually yielding oxidation derivatives. The most prominent of the latter is homogentisic acid. This research has shown that this acid is present in unstim-

<sup>11</sup> THIELE, R., Die Verarbeitung des atmosphärischen Stickstoffs durch Mikroorganismen. Landw. Versuchs-Stat. **63**:161-238. 1906.

<sup>12</sup> DE VRIES, HUGO, Die Neuzüchtungen Luther Burbank's. Biol. Centralbl. **26**:609-621. 1906.

<sup>13</sup> CZAPEK, FRIEDRICH, unter Mitwirkung von RUDOLPH BERTEL, Oxydative Stoffwechselvorgänge bei pflanzlichen Reizreaktionen. (Zwei Abhandlungen.) Jahrb. Wiss. Bot. **43**:361-467. 1906.

ulated tips in about the quantity of 16 per cent., while in stimulated tips it is present to the extent of about 20 per cent. Such figures can be obtained with considerable accuracy by an improved application of the silver reduction test. The increased reduction of an ammoniacal solution of silver nitrate following stimulation (geotropic or phototropic) can be positively shown to be due to the increased quantity of homogentisic acid. The oxidation of this acid is accomplished in ordinary metabolism by an enzyme, which because of its activity with phenol is called phenolase. The normal activity of this enzyme prevents accumulation of homogentisic acid above c. 16 per cent. In consequence of stimulation, however, there arises a substance (*de novo* so far as our knowledge now goes) which inhibits the activity of the enzyme, and thus the accumulation of homogentisic acid up to 20 per cent. is indirectly permitted. This new substance is called anti-enzyme. For different organs of the same individual and for different individuals of close systematic relationship this anti-enzyme is identical. With decreasing relationship, however, the actual substance of the anti-enzyme seems to differ, though the function remains constant. Chloroform narcosis, suffocation, antipyrin, acids, alkalies, various traumatic influences, mechanical interference with growth, and universal illumination all gave negative results for an anti-enzyme. Thus the author believes that this increased reduction of silver following stimulation is a safe and reliable test for tropistic sensation.—RAYMOND H. POND.

**Behavior of liverworts in darkness.**—NĚMEC has been experimenting with certain bryophytes which will grow in darkness and are not geotropic, to see what light they can throw on the mode of perception of geotropic stimulus, the biological significance of etiolation, and spontaneous nutations.<sup>14</sup> He finds different species behave very differently in darkness; some do not grow, others make a little growth but show no etiolation, while yet others grow long and vigorously and become markedly etiolated. Of the last, most are geotropic and grow erect or obliquely upward. But *Lejeunea bidentata* and *L. serpyllifolia* are ageotropic, having no starch or other statoliths. In darkness their shoots are completely unoriented, being at first hyponastic and then nutating irregularly. The same disorientation is observable in the sporogone of *Aneura pinguis*, but the vegetative shoots are geotropic and contain abundant statolith starch. The sporogone of *Pellia calycina* behaves similarly, but is slightly geotropic at first, losing this during elongation of the seta, which contains still some starch diffused between cell wall and vacuole and of slight mobility. The sporogone of *Pellia epiphylla*, on the contrary, is strongly geotropic and has abundant mobile starch. The sporogones of the three last named are strongly positively phototropic, and the reaction is in no way connected with the capsule. Also the upper part of the seta may be cut off, neither wound shock (which is transient) nor removal interfering with the reaction. The vegetative shoots of *Pellia calycina* grow well (plagi-

<sup>14</sup> NĚMEC, B., Die Wachstumsrichtungen einiger Lebermoose. Flora 96:409-450. figs. 9. 1906.

otropically) in darkness, and give a very characteristic geotropic reaction, having abundant statolith starch. *P. epiphylla* generally does not grow in darkness. Neither these two nor *Lophocolea bidentata* are geotropic in light. The behavior in darkness of *P. epiphylla*, the two species of Lophocolea, and the sporogone of *Aneura* must be considered as purposeless.

The researches appear to strengthen the statolith theory of geotropic perception.—C. R. B.

**Photosynthesis.**—USHER and PRIESTLY, having shown in a previous paper<sup>15</sup> that CO<sub>2</sub> may be decomposed in plants independently of enzymic or "vital" activity provided the products (H<sub>2</sub>O<sub>2</sub> and H'COH) are removed, have now devised an arrangement by which photolysis of CO<sub>2</sub> can be produced *in vitro*.<sup>16</sup> By covering gelatin plates, or even water, with a very thin uniform layer of chlorophyll deposited from solution, and placing the glass plate or the shallow dish of water carrying the film in a vessel with CO<sub>2</sub> and illuminating it, they found formaldehyde produced and were able to recover identifiable quantities. By making up the gelatin with an aqueous solution of a catalase, the hydrogen peroxid also produced was decomposed with the evolution of O<sub>2</sub>, 2cc being obtained in one case. The chlorophyll was gradually bleached, in harmony with the view that it acts as a sensitizer and is destroyed in the process of photolysis. Synthesis of H'COH into carbohydrate was found in the earlier paper to be dependent on the healthy condition of the protoplast, and feeding experiments have already shown that H'COH when supplied in very dilute form can be condensed by green plants if illuminated. The authors painted the white petals of *Saxifraga Wallacei* with chlorophyll, and floated them on water charged with CO<sub>2</sub> in light. In the course of a day they were found to contain starch. Thus they secured photosynthesis by a different (though inefficient) arrangement of the mechanism of a green leaf.

Further study was made of photolysis of CO<sub>2</sub> in the presence of uranium salts. They have direct evidence of the production of formic acid (which also is produced under some conditions in the plant as an intermediate product in the reduction of CO<sub>2</sub>), but they were unable to isolate and identify formaldehyde.

These papers record a most important step in solving the problems of photosynthesis.—C. R. B.

**Vascular anatomy of cycads.**—Two years ago MATTE published his thesis on the vascular anatomy of the cycads,<sup>17</sup> in which he presented in great detail, with copious illustrations, the vascular anatomy of the leaves and flowers in representative species of all the nine genera; and of the seedlings of *Dioon edule*, *Cycas*

<sup>15</sup> USHER, F. L., and PRIESTLY, J. H., A study of the mechanism of carbon assimilation in green plants. Proc. Roy. Soc. London B. 77:369-376. 1905.

<sup>16</sup> ——The mechanism of carbon assimilation in green plants: the photolytic decomposition of carbon dioxid *in vitro*. *Idem* 78:318-327. 1906.

<sup>17</sup> MATTE, H., Recherches sur l'appareil libéro-ligneux des Cycadées. pp. 233. pls. 16. figs. 264. 1904.

*siamensis*, and *Encephalartos Barteri*. He lays special stress on the strands of the foliar organs, his unit of primary importance in relation to phylogeny being what he calls the *meriphyte*, the vascular axis of the stem being of secondary importance. Among other conclusions in reference to the strands of the leaves, he states (1) that the course of the vascular strands sometimes presents great complexities, and (2) that the classic Ω of the foliar arc, although usual, is not general, referring particularly to *Bowenia*. A later paper<sup>18</sup> deals with the structure of the meriphyte of *Bowenia spectabilis*, and attempts to prove that the complex arrangement of the foliar strands even in this form can be reduced to the typical Ω.—R. THIESSEN.

In an earlier paper MATTE<sup>19</sup> noted in certain ovulate sporophylls of *Ceratozamia* the occurrence of three or more vascular strands near the abaxial surface of the sporophyll, in addition to the row of strands that usually appears in transverse section. These accessory strands showed reversed orientation, the xylem being on the abaxial side; and MATTE regards this as an evidence of the union of two sporophylls along their edges.—CHARLES J. CHAMBERAIN.

**Problems of nutrition.**—ARTARI published in 1904 an account of the influence of the different concentrations of glucose and saccharose upon the development of *Stichococcus bacillaris* and certain lichen-algae. This he now supplements<sup>20</sup> by investigating the effect of different concentrations of each component of the nutritive solutions upon the amount and energy of multiplication in *Stichococcus Chlorella* (a new physiological species) and the algae from *Xanthoria parietina*.

The concentration of  $\text{NH}_4\text{NO}_3$ , which was the source of N, has no effect on Chlorella below 0.5 per cent., and on *Stichococcus* below 1 per cent.; but higher concentrations retard development, and suggest that the salt acts also in another way than by its osmotic pressure. The nutritive value of various sources of N varies according as glucose is present or absent. Glucose itself exercises an observable effect even at 0.005 per cent., and acts best between 0.5 and 2 per cent. The limiting concentration of monosaccharides, varying more or less with different algae, seems to be related to molecular weight, since it is about equal in monosaccharides (glucose, galactose, and fructose), and approximately double this limit in disaccharides (saccharose and lactose), which also agree together. Many other details, not easily related, are also recorded. A few observations upon the effect of concentration upon the form and size of cells are given incidentally; the author seems not to know of LIVINGSTON's papers on this subject.—C. R. B.

<sup>18</sup> MATTE, H.; Compléments à la structure mérophytaire du *Bowenia spectabilis*. Compt. Rend. Acad. Sci. Paris, 409-416. 1905.

<sup>19</sup> MATTE, H., Une anomalie de structure dans l'écailler ovulifère de *Ceratozamia mexicana*. Bull. Soc. Linn. Normandie V. 7:52-54. 1903.

<sup>20</sup> ARTARI, A., Der Einfluss der Konzentrationen der Nährlösungen auf die Entwicklung einiger grüner Algen. II. Jahrb. Wiss. Bot. 43:177-214. 1906.

**Tetrads of a Drosera hybrid.**—ROSENBERG<sup>21</sup> states that in the genus *Drosera* the pollen cells remain united in tetrads at maturity, even after pollination. In the hybrid *D. rotundifolia*  $\times$  *D. longijolia*, in which he previously studied the earlier stages of pollen development,<sup>22</sup> he now describes the mature stage after the reduction division. Instead of four pollen cells in such a tetrad, he finds occasionally in the hybrid additional small pollen grains, which arise from extra nuclei formed by chromosomes left behind in the cytoplasm during the reduction divisions. The tetrad groups of the parent species are distinguishable by the size and shape of their individual grains, those of *D. rotundifolia* being smaller and rounded at the outer edge, those of *D. longijolia* larger and flattened. In most of the tetrads of the hybrid containing only four pollen cells, all the grains have the size and shape of *D. longijolia*; but occasionally two cells have the characters of one parent and two those of the other. He concludes that in the distribution of chromosomes in the reduction divisions, chromosomes from both parents usually enter all the daughter nuclei, but that occasionally in the first division the chromosomes of *D. rotundifolia* are more or less completely segregated from those of *D. longijolia*, two of the pollen cells of such a tetrad having the characters of the pollen of each parent.—R. R. GATES.

**Items of taxonomic interest.**—S. H. BURNHAM (Torreya 6:235. 1906) has described a new *Monotropsis* from the Blue Ridge Mountains.—W. H. BLANCHARD (*idem* 236) has described a new *Rubus* (dwarf blackberry) from Vermont.—B. F. BUSSE (Rept. Mo. Bot. Garden 119-125. 1906) has described new Texan species under *Tracyacanthus*, *Allium*, *Psoralea* (2), *Tragia*, *Xanthium*, *Antennaria*, and *Silphium*.—A. S. HITCHCOCK (Rhodora 8:205-212. 1906), among notes on grasses of the N. E. United States, describes new species in *Panicum* (4) and *Glyceria*.—W. H. BLANCHARD (*idem* 212-218) describes 2 new species of *Rubus* (blackberries) from Maine.—A. BERGER (Notizblatt 4:250. 1906) has described a new *Agave* from Mexico.—H. H. HAINES (Jour. Linn. Soc. 37:407-409. 1906) has described 2 new species of *Populus* from India.—O. STAPP (*idem* 495-532), among descriptions of numerous new species from the Dawe's collection from Uganda, publishes the new genus *Blasamocitrus* (Rutaceae).—J. HUBER (Boletim Mus. Goeldi 4:510-619. 1906), in his sixth contribution to a flora of the Amazons, describes *Browniopsis* as a new genus of Leguminosae (Caesalpinoideae). J. C. ARTHUR (Journal of Mycology 13:28-32. 1907) has published *Polioma*, *Spirechina*, *Prospodium*, and *Nephlyctis* as new genera of Uredinales.—PH. VAN TEIGHEM (Ann. Sci. Nat. Bot. IX. 4:223-260. 1906) has established the new dicotyledonous family Agialidaceae, to include certain spiny trees and shrubs referred usually to *Balanites* (Simarubaceae), the

<sup>21</sup> ROSENBERG, O., Erblichkeitsgesetze und Chromosomen. Botaniska Studier, tillägnade F. R. KJELLMAN 1906:237-243. figs. 5.

<sup>22</sup> —, Das Verhalten der Chromosomen einer hybriden Pflanze. Ber. Deutsch. Bot. Gesells. 21:110. 1903.—Über die Tetradentheilung eines Droserabastardes. Ber. Deutsch. Bot. Gesells. 22:47. 1904.

family containing the genera *Balanites*, *Agialida*, and *Agiella* (new); the same author (*idem* 261-271) also breaks up the Boraginaceae into 4 families, Boraginaceae, Ehretiaceae, Cordiaceae, and Heliotropiaceae.—C. V. PIPER (Contrib. U. S. Nat. Herb. 10:1-48. pls. 1-15. 1906), in a revision of N. Am. species of *Festuca*, recognizes 34 and describes 3 as new.—A. A. HELLER (Muhlenbergia 2:177-256. 1906), in an account of botanical exploration in California during 1906, in which numerous species are noted and nomenclatural changes made, describes new species in *Taxicoscordion*, *Eriogonum* (2), *Mirabilis*, *Lupinus* (4), *Hesperastragalus*, *Anogra*, *Chylisma*, *Phlox*, *Phacelia*, *Conanthus*, *Cryptantha* (2), *Amsinckia*, *Pentstemon* (2), and *Orthocarpus*.—PHILIP DOWELL (Bull. Torr. Bot. Club 33:547-556. pls. 18-22. 1906), in a revision of N. Am. species of *Calceolaria*, recognizes 16 and describes 6 as new.—J. M. C.

**Formative influence of light.**—PEIRCE has extended his studies on irritability in plants from algae to liverworts and ferns, recording the effect of light upon germination and early growth in *Anthoceros fusiformis*, *A. Pearsoni*, *Fimbriaria californica*, and *Gymnogramme triangularis*, and its effect on form in later stages of growth.<sup>23</sup> He finds that germination is dependent upon light of undetermined intensity, and that direction of growth and of successive cell division is determined by the direction of light. The direction of light also profoundly modifies the form of the thalli of the liverworts. This was most marked in *Anthoceros*. On a clinostat they tend to become solid, erect, and cylindrical, conic or vasiform, with radial structure instead of dorsiventral. The author holds that his early results support the hypothesis that, aside from what is actually transmitted from parents to offspring, likeness is due to likeness or identity of the physical environment, and that these factors are as essential determinants as the substance transmitted. Unfortunately PEIRCE's experiments were interrupted by the earthquake of April 16, so that conclusive tests have not been made in all cases. The work will be continued.—C. R. B.

**Assimilation of organic acids by algae.**—An interesting contribution to our knowledge of the power of assimilation of carbon compounds by autotrophic plants has been made by TRÉBOUX.<sup>24</sup> This investigator experimented with some forty species of the lower algae, testing their power of assimilating various organic acids, which were given in the form of potassium or ammonium salts. Cultures were kept in absolute darkness. It was found that about one-half of the species flourished under the cultural conditions with an organic acid as the only source of carbon; and that of all the acids used acetic acid was most readily assimilated, while acids with larger carbon chains were assimilated in only a few cases. In some cases aminoacids were used with accompanying escape of ammonia. This

<sup>23</sup> PEIRCE, G. J., Studies of irritability in plants. Annals of Botany 20:449-465. pl. 35. 1906.

<sup>24</sup> TRÉBOUX, O., Organische Säuren als Kohlenstoffquelle bei Algen. Ber. Deutsch. Bot. Gesells. 23:432-441. 1906.

work contributes additional evidence showing that the line between autotrophic and heterotrophic nutrition is not so sharply drawn as is generally supposed. The power of the lower algae to use carbon compounds and especially the lower acids explains the abundant growth of those forms where decaying organic matter is present, as in contaminated waters.—H. HASSELBRING.

**Rhizomorphic root-rot of vine.**—MAGNUS<sup>25</sup> describes a rhizomorphic root-rot of the vine which causes the death of numerous plants in the vineyards of certain regions of Europe. The rhizomorphs are white strands which may attain a diameter of 2<sup>mm</sup>. The strands lack a well-developed cortex, thereby differing from the characteristic strands of *Armillaria mellea*. The fungus is found to be abundant on the posts used as supports for the vines. From the posts the strands spread to the young roots of the vine, entwining and killing them. Although this disease with the accompanying rhizomorphs had been known for several years, it was not until the present year that the connection of the rhizomorphs with one of the Hymenomycetes was established by finding numerous sporophores of *Collybia platyphylla* growing from the strands. As this fungus is very common in America, it is not unlikely that it may be connected with one of the numerous rhizomorphic root-rot diseases infecting our fruit trees.—H. HASSELBRING.

**Segregates of *Rhus glabra*.**—GREENE<sup>26</sup> has studied the forms referred to *Rhus glabra* throughout its reputed range. He remarks that "there is no one species of tree or shrub of any continent that really holds the geographic range which the books and lists ascribe to *Rhus glabra*;" and points out the exceedingly diverse areas it is said to occupy in its continental distribution. Accordingly he has begun its segregation on the basis of such herbarium material as is available, recognizing the fact that this is probably a very scanty showing of the real situation, for "no special call has been made for collecting these shrubs from different regions." The form chosen to stand for the original *R. glabra* L. is one ranging from eastern Virginia and southern Maryland through southern Pennsylvania to Connecticut. Outside of that region botanists will have to refer their reputed forms of *R. glabra* to other species. This initial work of segregation has resulted in 29 species, 24 of which are new.—J. M. C.

**Transpiration.**—ARESCHOUW maintains the correctness of his view that the palisade tissue, when well developed and compact, reduces transpiration,<sup>27</sup> explaining away certain apparently contradictory experiments of HESSELMAN and others.

It seems to be about time to dismiss the idea of transpiration as a function,

<sup>25</sup> MAGNUS, P., Ueber eine Erkrankung des Weinstockes. Ber. Deutsch. Bot. Gesells. 24:402-406. 1906.

<sup>26</sup> GREENE, EDWARD L., A study of *Rhus glabra*. Proc. Wash. Acad. Sci. 8: 167-196. 1906.

<sup>27</sup> ARESCHOUW, F. W. C., Ueber die Bedeutung des Palisadenparenchym für die Transpiration der Blätter. Flora 96:329-336. 1906.

of which leaves are the organs, and to consider it as a merely unavoidable evaporation whose amount depends upon physical factors that need to be exactly evaluated. What would be thought of an engineer who attempted to compare the performance of two boilers of different construction if he knew nothing of the heat units applied? Yet most of the so-called "comparative" studies of transpiration take no account of the fundamental energetics involved, assuming that when leaves are placed "under the same conditions" they have a like amount of energy for transpiration!—C. R. B.

**Correlation and leaf size.**—A. J. EWART has published a short note dealing with correlation and leaf size.<sup>28</sup> LINDEMUTH has shown that adult leaves of *Begonia* and *Iresine* increase in size when allowed to root in the soil. EWART conducted experiments on *Tilia europaea*, and found that adult leaves do not increase in size, even when most of the leaves are removed. However, a partial defoliation of young shoots causes a development of unusually large leaves, and the increase in size beyond the normal is due to an increase in the number of cells and not to an increase in their size, as supposed by LINDEMUTH. That increased size is due to an increased number of cells in such cases is not the common view.—H. C. COWLES.

**Presynapsis and synapsis.**—Presynaptic and synaptic stages in the first division of the embryo sac mother cell of *Adoxa* are interpreted by LAGERBERG as follows.<sup>29</sup> Before synapsis the chromatin granules collect into groups which are often in pairs, apparently less numerous than the mature chromosomes. The chromatic substance of the groups becomes distributed along parallel linin threads, so that at the beginning of synapsis there are two parallel threads which fuse as synapsis proceeds. The single thread which is thus formed is the thickest which appears at any time in this nucleus. Accordingly, the fusion of the male and female elements of the nucleus takes place during synapsis.—CHARLES J. CHAMBERLAIN.

**Embryology of Capsella.**—The late Mrs. Mabel Schaffner<sup>30</sup> left an incomplete paper on the embryology of *Capsella*, which her husband, Professor JOHN H. SCHAFFNER, has published. It is a detailed study of the development of the embryo of what is perhaps the most frequently used dicotyledon in teaching. Aside from the completeness of the series, the striking feature of the plates is that the whole series is drawn to the same scale, representing to the eye the actual increase of the embryo in size at each stage of its development.—J. M. C.

<sup>28</sup> EWART, A. J., The influence of correlation upon the size of leaves. *Annals of Botany* 20:79-82. 1906.

<sup>29</sup> LAGERBERG, TORSTEN, Ueber die präsynaptische und synaptische Entwicklung der Kerne in der Embryosackmutterzellen von *Adoxa moschatellina*. *Botaniska Studier, tillägnade F. R. KJELLMAN*. 1906:80-88.

<sup>30</sup> SCHAFFNER, MABEL, The embryology of the shepherd's purse. *Ohio Nat. 7:1-8. pls. I-3.* 1906.

**Leaf structure.**—Most botanists are familiar with the contributions of Dr. COCKAYNE to our knowledge of the flora of New Zealand and the adjacent islands. On his trip to the southern islands several years ago, he brought back a number of living plants which are now growing in the rockery of Canterbury College. These plants have been made the subject of an anatomical investigation by Miss HERRIOTT.<sup>31</sup> In spite of the uniform climate of the southern islands, many if not most of the plants reveal xerophytic structures. The author cites two conditions in these island regions which may be regarded as xerophytic. One is the frequency and violence of the wind, and the other is the peaty soil.—H. C. COWLES.

**Forestry.**—Two recent bulletins of the Bureau of Forestry will be of some interest to ecological workers. One of the bulletins<sup>32</sup> describes the valley and ridge forests of Kansas and Nebraska and their economic possibilities. The natural extension of both types of forest is discussed, but this subject will be well known to all through work of Professor C. E. BESSEY. The other bulletin<sup>33</sup> is almost wholly of economic interest, but ecologists will find some notes on the natural reproduction of the white pine in the old fields of New England.—H. C. COWLES.

**Hygroscopic mechanisms.**—STEINBRINCK, who has given particular attention to hydrophysics, summarizes for non-technical readers the mechanisms of shrinkage and cohesion in plants.<sup>34</sup> It is these mechanisms which are concerned chiefly in the rupture of capsules and sporangia. The current conceptions as to the rupture of the latter particularly need correction, and the article is commended to students. In his *Literaturübersicht* he has indeed overlooked American literature on this subject.—C. R. B.

**Biology of Dunaliella.**—In 1905 TEODORESCO described<sup>35</sup> *Dunaliella* as a new genus of Volvocaceae, giving an account of its structure and life history. In a second paper<sup>36</sup> he has begun an account of certain "biological observations." In this first instalment these observations deal with changes in the form of the body, cell structure, cell division, sexual reproduction, and the resting condition.—J. M. C.

<sup>31</sup> HERRIOTT, E. M., On the leaf structure of some plants from the southern islands of New Zealand. *Trans. N. Z. Inst.* 38:377-422. 1906.

<sup>32</sup> KELLOGG, R. S., Forest belts of western Kansas and Nebraska. U. S. Dept. Agric., Forest Service, Bull. 66. 1905.

<sup>33</sup> SPRING, S. N., The natural replacement of white pine on old fields in New England. U. S. Dept. Agric., Bureau of Forestry, Bull. 63. 1905.

<sup>34</sup> STEINBRINCK, C., Ueber Schrumpfungs- und Kohäsions-mechanismen von Pflanzen. *Biol. Centralbl.* 26:657-677, 721-744. figs. 28. 1905.

<sup>35</sup> Beih. Bot. Centralbl. 18:215-232. 1905.

<sup>36</sup> TEODORESCO, E. C., Observations morphologiques et biologiques sur le genre *Dunaliella*. *Rev. Gén. Botanique* 18:353-371. pls. 2. 1906.

Purple bacteria.—MOLISCH describes two new genera of purple bacteria isolated in the laboratory from jars of sea water, sea grass, and decaying crabs or star-fish.<sup>37</sup> For these forms he finds it necessary to add a new sub-family to WINOGRADSKY's and MIGULA's classification. This is sub-family VI. RHODOCAPSACEAE (cells free, not capable of swarming throughout life). Under this he places his two new forms, *Rhodocapsa suspensa* (cells sometimes capable of swarming, rods or threads with gelatinous capsule), and *Rhodothece pendens* (power of swarming not as yet observed, cells round, with gelatinous capsule).—MARY HEFFERAN.

Origin of the pollen tube.—According to WETTSTEIN,<sup>38</sup> the rhizoidal tubes of the cycads and Ginkgo are to be regarded as completely homologous with the pollen tubes of the higher gymnosperms and the angiosperms. The writer then attempts to show that the rhizoidal tube of the cycads and Ginkgo is homologous with the vegetative end cell of the heterosporous pteridophytes. The rudimentary or rhizoidal cell of the heterosporous pteridophytes does not lead to the rhizoidal tube of the cycads and Ginkgo, and is still represented in these lower gymnosperms by a prothallial cell. Chalazogamy is regarded not as a primitive condition, but merely as an interesting special case. The paper is a preliminary statement without figures.—CHARLES J. CHAMBERLAIN.

<sup>37</sup> MOLISCH, HANS, Zwei neue Purpurbakterien mit Schwebekörperchen. Bot. Zeit. 64:223-232. pl. 8. 1906.

<sup>38</sup> WETTSTEIN, R. VON, Der Ursprung des Pollenschlauches. Naturw. Rundschau 21:1-2. 1906.

## NEWS

DR. F. CAVARA has been appointed director of the Botanical Garden at Naples.

DR. LUDWIG DIELS, University of Berlin, is delivering a series of lectures at the University of Marburg.

DR. B. LONGO, lately of the University of Rome, has been appointed professor of botany at the University of Siena.

DR. ARTHUR HOLICK has been appointed by the mayor a member of the Board of Education of Greater New York.

THE AMERICAN ASSOCIATION for the Advancement of Science will meet at the University of Chicago during the holiday week of 1907.

DR. J. B. FARMER has been appointed president of the section of botany of the British Association, which holds its next meeting at Leicester.

DR. D. H. SCOTT has resigned his position as honorary keeper of the Jodrell Laboratory at Kew, a position which he has held since 1892.

PROFESSOR C. E. BESSEY, University of Nebraska, has been elected vice-president of Section G of the American Association for the coming year.

MISS CLARA E. CUMMINGS, professor of cryptogamic botany in Wellesley College, died on December 28. She had served Wellesley College since 1878.

A. G. TANSLEY, assistant professor of botany, University College, London, and editor of the *New Phytologist*, has been appointed lecturer in botany to succeed Professor A. C. SEWARD.

MISS LILIAN M. WILCOCKSON, of the University of Minnesota, is arranging a "science and recreation camp" on the shore of Vermilion Lake, Minnesota, to begin about the middle of July. Work and play are offered *ad libitum*.

ABOUT HALF THE COLLECTIONS have been transferred from the old building in Berlin to the new Museum building of the Royal Botanic Garden at Dahlem. It is expected that the transfer will be complete by spring and the building then fully occupied.

THE HERBARIUM of the late WILLIAM MITTEN of England, one of the most important collections for American bryology, has been purchased by the New York Botanical Garden. We congratulate the director upon his enterprise and success in securing this valuable collection.

AT THE New York meeting of the Botanical Society of America the following officers were elected for the ensuing year: President, GEORGE F. ATKINSON, Cornell University; Vice-president, N. L. BRITTON, New York Botanical Garden; Secretary, D. S. JOHNSON, Johns Hopkins University; Treasurer, ARTHUR HOLICK, Columbia University.

# BOTANICAL GAZETTE

MARCH 1907

RUBIACEAE: ANATOMICAL STUDIES OF NORTH AMERICAN REPRESENTATIVES OF CEPHALANTHUS,  
OLDENLANDIA, HOUSTONIA, MITCHELLA,  
DIODIA, AND GALIUM

THEO. HOLM

(WITH PLATES VII-IX)

To students of plant anatomy few books have proved to be of greater service than SOLEREDER'S *Systematische Anatomie der Dicotyledonen*.<sup>1</sup> It is a book that contains a vast amount of information as to the work that has been accomplished in anatomy thus far, and gives in concise and clear form the most important results in respect to the general characters of the dicotyledonous families, as well as a number of purely generic or even specific peculiarities observed in their internal structure; the classified lists of the enormous literature bearing on this subject are of no less importance. Whether the book be consulted for the sake of ascertaining the distinguishing characters of certain families or genera, or for obtaining a general view of the structure possessed by the dicotyledons, one is always sure to obtain exact information as to the plants that have been treated from this particular point of view. The number of dicotyledons that have thus far been studied is of course very large, and very few of the most interesting families or genera have escaped the attention of investigators. Most of the parasitic, saprophytic, and climbing plants, with their more or less anomalous structure, have already been duly considered and very carefully described.

However, when we remember that most of the literature has been contributed by European botanists, it is readily understood that very

<sup>1</sup> Stuttgart, 1899.

many of the extra-European plants have been studied either from specimens cultivated in botanical gardens or preserved in herbaria. This may be the cause why, for instance, the root-structure has not been studied to the same extent as that of the other organs, for the root system and rhizomes are not satisfactorily represented in herbaria, or may be entirely wanting. Also, only a very few of the more common genera and species of the North American flora have been studied anatomically, because European botanists have not been in a position to study the American plants, and because American botanists have not paid sufficient attention to anatomical botany. American students, therefore, have an important and interesting task before them in undertaking an anatomical investigation of their native plants, the common as well as the rarer ones, in order to assist in the completion of a work so well begun and so very instructive as that of SOLEREDER and his predecessors.

Moreover, the study of plant structures is necessary to the fuller understanding of ecology. The plant societies, so excellently outlined by WARMING and SCHIMPER, should not be determined merely by the social occurrence of a number of types that characterize a certain vegetation, but they should be investigated much farther; thus we might be able to distinguish between characters that may be looked upon as those of the family and those that are purely epharmonic. Very frequently, so far as we know, these structural characters do not correspond with what might be expected from the nature of the habitat. Halophytes and xerophytes are often not to be separated by means of their structure alone; for example, many bog plants exhibit peculiarities that are familiar to us as xerophytic, and *vice versa*. If the structure of all the most significant components of these societies was so well understood that we were able to distinguish between epharmonic characters and those that are generic or specific, we might gain a clearer idea of the real factors that have brought these plants together so as to form societies.

Another and perhaps more important problem is the application of the structures as a means of distinguishing genera and species; in other words, to bring together such points of distinction as may be observed in both the external and internal morphology of plants. For this purpose almost any contribution, large or small, may be of

some service; and the writer is under the impression that a detailed account of a few plants, hitherto left unstudied, may prove more useful than a broad anatomical treatment of a number of genera and species more or less vaguely described or insufficiently compared.

Among the North American plants that have not been studied fully thus far are the Rubiaceae, at least the genera enumerated in the title of this paper. With the object of presenting a contribution to the knowledge of some of these plants, the writer has endeavored to gather as many data as possible from the vegetative organs which may be of some interest to students of plant anatomy. The following species have been studied:

*Cephalanthus occidentalis* L. (swamps near Brookland, D. C.), *Oldenlandia glomerata* Michx. (swamp near Brookland, D. C.), *Houstonia coerulea* L. (open thickets, D. C.), *H. purpurea* L. (with the preceding), *Mitchella repens* L. (wooded ravines near Sligo, D. C.), *Diodia teres* Walt. (open fields, Brookland, D. C.), *Galium pilosum* Ait. (thickets, Brookland, D. C.), *G. triflorum* Michx. (woods near Anacostia, and on the Potomac shore, Va.), *G. circaeans* Michx. (with the preceding), *G. latifolium* Michx. (Biltmore, N. C., the specimens kindly furnished by Mr. C. D. Beadle).

#### CEPHALANTHUS OCCIDENTALIS (NAUCLEAEAE B. et H.)

**The root.**—In small shrubs, too young to produce flowers, the primary root persists and is quite large; it is of a brownish color and measures about 1.5<sup>cm</sup> in thickness at the base. At a depth of about 9<sup>cm</sup> it commences to branch, dividing into a few slender, very long branches. Numerous white lateral roots develop on all sides, which are very hairy and branch freely. A lateral root of first order shows the following structure. Inside the epidermis is an exodermis (*fig. 1, ex*) of thin-walled, pentagonal cells which covers a stratum of several cell-layers, a tissue representing cork (*fig. 1, p*). The cortical parenchyma consists of ten strata of thin-walled cells arranged radially and with very wide intercellular spaces, sometimes wide enough to be called lacunae. Neither starch, crystals, nor raphides were observed in the cortex. The innermost layer of the cortex is differentiated as a thin-walled endodermis with the Casparyan spots plainly visible. A thin-walled pericambium surrounds the leptome and hadrome, the primitive structure of which could not be ascertained since secondary tissues had already become developed; the

conjunctive tissue is quite thick-walled, but represents only a minor portion of the central cylinder.

In the capillary lateral roots of second order, in which no increase in thickness has taken place, there is only one tangential division of the phellogen and only three layers of cortex. The root is hexarch, six groups of leptome alternating with six rays of wide vessels, and with a central group of thick-walled conjunctive tissue. The development of a phellogen has thus taken place in these roots before the increase in thickness of the central cylinder has commenced.

The formation of cork inside the exodermis seems to be a point of interest. In roots of dicotyledons the cork usually develops from the pericambium, and DEBARY<sup>2</sup> records only two cases (*Clusia* and *Bignonia capreolata*) in which the formation of cork takes place in the peripheral strata of the primary cortex, as described by VAN TIEGHEM.<sup>3</sup> A similar structure, however, has been detected in *Artanthe pothifolia*, *Jasminum humile*, and *Ruyschia Souroubea* by OLIVIER.<sup>4</sup> Since this peculiarity in regard to the superficial development of cork has been obscured in *Bignonia*, I examined also the roots of *Tecoma radicans* and found the same structure; thus Cephalanthus and Tecoma may be added as examples of this very rare structure of roots.

**The stem.**—In the flower-bearing shoots the leaves are generally in whorls of three, and the internodes become obtusely triangular. A smooth but thick cuticle covers the small-celled epidermis, which is moderately thick-walled, and inside of which several (about six) layers of cork are to be observed. Between the phellogen and the cortex proper is a broad continuous zone of collenchyma, which contains much chlorophyll. The cortical parenchyma is here differentiated into two very distinct zones: a peripheral, of about nine layers of large cells with wide, irregular lacunae; and an inner, of about eight compact strata of smaller cells, interrupted here and there by small, isolated strands of typical stereome. Of these two zones the

<sup>2</sup> *Vergleichende Anatomie*, p. 563. 1877.

<sup>3</sup> *Recherches sur la symétrie de structure des plantes vasculaires*. Ann. Sci. Nat. Bot. V. 13:258. 1870.

<sup>4</sup> *Recherches sur l'appareil tégumentaire des racines*. Ann. Sci. Nat. Bot. VI. II:124. 1880.

peripheral contains chlorophyll, besides calcium-oxalate in the shape of crystalline sand. Inside the cortex is the central cylinder, consisting of a broad zone of leptome, some strata of cambium, and many rows of vessels separated by narrow medullary rays. The central portion of the internode is occupied by a thin-walled but solid pith, in which crystalline sand was noticed.

A somewhat modified structure is to be observed in the shoots of younger specimens, which are yet purely vegetative. In these the leaves are opposite, not whorled in threes as in the floral shoots, and the outline of the internodes is cylindric. The cuticle is wrinkled, and no phellogen is developed; thus the collenchyma borders directly on epidermis. The differentiation of the cortex into two zones is not so distinct, since the parenchyma is more uniformly developed, more compact, and with the cells radially arranged from the periphery to the stele, except where it is interrupted by the stereome, which occurs in very small, scattered groups. There is no endodermis, and the structure of the mestome and pith is identical with that of the floral shoots.

In comparing these internodes of floral and vegetative shoots with the slender peduncle that bears the globose inflorescence, it is interesting to notice that the structure is almost identical, with the exception that the number of strata of the various tissues is smaller, that no cork is developed, and that bicellular pointed hairs abound.

**The leaf.**—The blade shows a bifacial structure. The cuticle is somewhat irregularly thickened so as to form striations, especially lengthwise (parallel with the midrib). Very characteristic is the small-celled epidermis with the radial walls straight on both faces of the blade; the stomata, which are confined to the lower face, have mostly two subsidiary cells parallel with the stoma (fig. 2), but variations are frequent, there being sometimes two subsidiary cells on one side of the stoma and only one on the other, or the differentiation of the epidermal cells surrounding the stoma may be so slight that no typical subsidiary cells are observable. The stomata are level with epidermis and the air chamber is quite wide and deep. While the upper surface of the blade is glabrous, the lower is hairy with uni- and bicellular hairs, which are long, more or less curved, with the apex acute, thick-walled, and covered by a striately thickened cuticle.

The chlorenchyma is differentiated into a typical and very compact palisade tissue of two strata, and a pneumatic tissue of oblong cells (in transverse section) more or less vertical to the blade and with very wide intercellular spaces. The mechanical tissue occurs only as collenchyma, which follows the veins and which is best developed on the leptome side. A small isolated mass of this tissue is also found in the margins of the leaf-blade. The collenchyma is quite thick-walled and is located directly inside the epidermis, but only as isolated masses above and below the nerves, besides in the margins. A thin-walled, colorless tissue, which evidently represents a water-storage tissue, is amply developed in the midrib and the larger secondaries, where it surrounds the mestome completely. Crystalline sand was observed in some of the cells of this tissue.

The mestome strands are all collateral, but the arrangement of the mestome is somewhat different. The smallest veins show an orbicular outline in transverse sections, and they are surrounded by a very distinct parenchyma sheath; in these the hadrome is located exactly above the leptome. On the other hand, in the midrib or the secondaries the mestome is in the shape of an arch with the ends curved inward, whose concave face looks toward the upper face of the leaf. In this arch the leptome follows the lower face and takes part in the curvature of the end; thus actually the leptome becomes moved from the lower to the upper portion of the mestome bundle. The hadrome is represented by numerous but short rows of vessels arranged above the leptome in the middle of the arch, but beneath it at the ends.

**The petiole.**—The structure is exactly the same as that of the midrib in the blade. The single, very broad, and arch-shaped mestome strand is located in a mass of colorless parenchyma with crystalline sand, and possesses a support of well-developed collenchyma, while chlorenchyma is completely absent.

The characteristics of *Cephaelanthus*, therefore, are the location of the phellogen inside the exodermis of the root; the absence of stereome in the leaf; the differentiation of the cortex of the stem into two distinct cones interspersed with stereome; and finally the structure of the larger veins in the leaf with the arch-shaped mestome strand.

## OLDENLANDIA GLOMERATA (HEDYOTIDEAE B. et H.)

An annual plant and the last of the Rubiaceae to bloom. It inhabits wet places on borders of swamps, being associated with *Hypericum multiflorum*, *Ilysanthes attenuata*, *Cyperus diandrus*, and *Fimbristylis autumnalis*, but is very rare in the District of Columbia. The short stem is erect from a decumbent base, and all the leaves, which are opposite and short petioled, subtend small cymose inflorescences; the apex of the stem is also terminated by an inflorescence. The basal internodes and the hypocotyl soon become bent towards the ground, and secondary roots develop freely from the full length of the internodes. There are buds in the axils of the cotyledons (one at each) which develop into small floral shoots of one or two internodes, but so late that their flowers are just beginning to open when those of the primary shoot are in fruit. The primary root is short, but has many lateral branches.

**The roots.**—The main root is hairy and has no exodermis. The cortex consists of three strata of very large, thin-walled cells, and the endodermis is thin-walled, with the Casparyan spots very distinct. The stele showed secondary formations in the leptome and hadrome, so that the primitive organization could not be ascertained. The lateral and secondary roots, on the other hand, do not increase in thickness and remain as diarch. They have long hairs, and show the same structure as the primary, but are much thinner. In the stele there are two strands of leptome, and the center is sometimes occupied by a wide vessel or by a small mass of conjunctive tissue. In these capillary roots the endodermis and pericambium are thin-walled and continuous.

**The stem.**—The hypocotyl is cylindric in contrast with the internodes above, which are quadrangular, and is perfectly smooth and glabrous. A thin-walled epidermis covers a cortex of three strata of very large cells in which raphides occur. The innermost layer is differentiated as an endodermis, which consists of small, thin-walled cells, surrounding the central cylinder. In this the leptome forms a closed ring around the hadrome, of which the rays are quite long, separated from each other by narrow medullary rays. A thin-walled pith occupies the center of the stele.

The upper internodes are obtusely quadrangular in cross-section,

with two slightly convex and two concave faces; pluricellular but relatively short hairs are to be observed along the angles. The cuticle is quite thick and shows longitudinal striations. The epidermis consists of rather large cells, with the outer wall somewhat thickened; there are several stomata of the same structure as those of the leaf, but the lateral walls of the subsidiary cells are straight, not undulate. Several of the epidermal cells contain large druids.

There is no collenchyma and no stereome, the cortex thus forming an uninterrupted cylinder around the stele. The cortex consists of six layers in the angles, and of four between them; it is a very thin-walled parenchyma of quite large cells with wide intercellular spaces. It contains chlorophyll, raphides, and druids of calcium oxalate. An endodermis of small, thin-walled cells, with the Casparyan spots very distinct, surrounds the stele of collateral mestome strands. In these the leptome forms an almost confluent zone, while narrow rays of parenchyma (one or two rows) separate the short hadromatic rays with only two or three vessels in each row. The greater portion of the central cylinder is occupied by the pith, which is thin-walled but solid, and in which druids are frequent.

Druids were thus observed in the parenchyma tissues as well as in the epidermis, and they are very conspicuous on account of their unusually large size.

**The leaf.**—The structure is bifacial, with the stomata confined to the lower face, and with the chlorenchyma differentiated into a typical pneumatic and a palisade tissue.

The cuticle is thin and smooth except where it covers the hairs and the midvein of the lower surface of the blade, where it shows a distinct wrinkling longitudinally. Viewed *en face* the lateral cell walls of the epidermis are undulate on both faces, and the stomata have two subsidiary cells, the lateral walls of which are frequently undulate like the others. Hairs are frequent above and below the larger veins; they are pluricellular, with mostly four cells in one row and the apical one pointed; the cell walls are rather thin. Along the margins of the blade the hairs are more frequent, but much shorter, consisting of only one or two cells. Viewed in transverse section, the leaf is perfectly smooth on the upper face, but on the lower the midrib forms an obtuse keel. The epidermis is thin-

walled on both faces, except below the midrib, where the outer walls become moderately thickened; a difference in regard to the lumen of the cells is very distinct, that of the ventral face being considerably wider than that of the dorsal. The stomata are slightly raised, and the air chamber is wide and deep.

A small water-storage tissue of two strata separates the epidermis from the parenchyma sheath of the median mestome strand on the dorsal face; on the ventral face this tissue is represented only by two or three cells underneath the epidermis and bordering on the parenchyma sheath.

The chlorenchyma, as already stated, is composed of two strata of palisade tissue on the ventral, and of pneumatic tissue on the dorsal. Of these the former is not quite typically developed, since the cells are rather low and broad. The pneumatic tissue is very open, the cells being irregular and branched, with wide intercellular spaces; cells with raphides are frequent in this tissue. There is neither collenchyma nor stereome in the leaves, and the parenchyma sheaths of the collateral mestome strands are thin-walled and the cells large. The midvein has a broad group of leptome and hadrome with narrow vessels, and is partly surrounded by the water-storage tissue.

**The stipules.**—The stipules in *Oldenlandia* are extended into two setae, which are terminated by a short, unicellular hair; while the margins bear a few (mostly three) much longer and pluricellular hairs, such as those described above from the stem. At the base of the petiole, but only on the ventral face, glandular hairs were observed. These have a very short stalk and resemble those of *Houstonia* (fig. 6); they are arranged in two small clusters, one on each side of the axillary bud, but without extending to the stipules.

The characteristics of the genus are the complete absence of collenchyma and stereome from the stem and leaves; the occurrence of very large druids of calcium oxalate in epidermis, cortex, and pith of the stem; also the pluricellular, pointed, and glandular hairs. But neither stem nor leaves indicate in their structure that the plant is a hydrophyte, any more than do the species of *Houstonia*.

#### *HOUSTONIA COERULEA* (HEDYOTIDEAE B. et H.)

The species is herbaceous, but perennial. The primary root is of short duration and soon becomes replaced by a number of secondary

roots which develop from the nodes (between the two leaves) and from the internodes. A small rosette of green leaves appears at the end of the first season, and this little shoot is the only portion of the seedling that winters over; it is developed from the axil of one of the basal leaves, and consists of a short, but very distinct, internode above ground and is terminated by a rosette of leaves, all opposite and with the internodes hardly perceptible. During the next spring an inflorescence becomes developed terminating the shoot; while at the same time numerous buds become visible in the axils of the leaves of the rosette, each producing a short decumbent stolon of one or two internodes, terminated by a dense rosette of green leaves. The stolons being above ground, all the leaves possess distinct petioles and blades. Secondary roots appear, as described above, at various places; they are very thin, of whitish color, and branch freely. The lateral shoot thus represents two stages of growth: a purely vegetative one during the winter, and a floral one during the next spring; while the primary shoot of the seedling blooms without being preceded by other leaves than those at the base of its own stem, including the cotyledons. Our species thus resembles an annual plant in blooming in the first year; a biennial in producing a rosette of leaves to winter over and to become terminated by an inflorescence in the next year; and finally it remains as a perennial by the continuous development of leafy shoots and inflorescences.<sup>5</sup> The inflorescence is a cyme of the dichasium type, but relatively few-flowered. These introductory remarks are to show that the roots are of short duration, that the stolons are above ground, and that the leaves are either cauline or basal, the latter constituting an over-wintering rosette.

**The roots.**—As stated above, the secondary roots are very slender; they are very hairy and the epidermis covers directly a cortical parenchyma of three, thin-walled, compact strata. An endodermis with the cell walls moderately thickened surrounds the central cylinder, in which the pericambium is continuous and thin-walled. These roots are diarch, the two rays of the hadrome meeting in the center, alter-

<sup>5</sup> A near ally of *H. coerulea* is *H. serpyllifolia* Michx., but in this species the vegetative shoots do not develop as rosettes of leaves, since all the internodes are stretched, with the leaves remote. In *H. rotundifolia* Michx. the habitus is much the same as in *H. serpyllifolia*, but the plant is more robust and the flowers are almost sessile and single in the leaf axils.

nating with two broad strands of leptome, and no increase in thickness takes place. A corresponding structure was observed in the capillary lateral roots, with the exception that the various tissues are much less developed, the cortex consisting of only two layers, the endodermis being thin-walled and the hadrome being reduced to a very few vessels.

**The stolons.**—The stretched internode is perfectly glabrous, and obtusely quadrangular to almost cylindric. A smooth, quite thick cuticle covers the epidermis, which is thick-walled and contains chlorophyll. There are four layers of compact cortical parenchyma with chlorophyll, and the endodermis is moderately thickened and contains starch. The stele is represented by a confluent zone of leptome and vessels in about five rows, with narrow medullary rays of mostly one row of cells. A thin-walled pith occupies the center and contains starch, but no raphides were observed.

**The aerial stem.**—The long internodes are glabrous, quadrangular, and four-winged. In these the cuticle is wrinkled, and the outer cell walls of the epidermis are quite thickened. Stomata are frequent; they are level with the epidermis and have a wide air chamber. The cortical parenchyma consists of four strata between the wings, but of eight in them; no mechanical tissue, neither stereome nor collenchyma, was observed. A thin-walled endodermis surrounds the collateral mestome strands and the pith, which is narrow and very thin-walled.

**The peduncle.**—There is here about the same structure as in the long internodes, but the outline is simply quadrangular. The cortex represents a narrower zone, and the stele consists of several more or less separate mestome strands, but in one circle and collateral as above; the medullary rays are very distinct and the central pith somewhat wider.

**The leaves.**—The basal leaves, which winter over, have a very distinct petiole and a blade that varies from spatulate to almost ovate. Viewed *en face* the blade shows a perfectly smooth cuticle, and the lateral cell walls of the epidermis undulate on both faces. Stomata occur only on the lower face (*fig. 7*), and they have a pair of subsidiary cells parallel with the stoma. Unicellular hairs with the cuticle spirally thickened (*fig. 5*) are to be observed along the margins, while bicellular hairs with similar spiral striations are distributed over

the upper face of the blade, especially near the apex. Viewed in transverse sections the blade shows a typical bifacial structure. The epidermis is thin-walled on both faces, but the lumen of the cells is considerably wider on the upper than on the lower face; the stomata are level with the epidermis. The chlorenchyma is differentiated into a palisade tissue of two strata and a pneumatic tissue of four layers; the latter tissue is very open and the cells are oblong to almost roundish, with wide intercellular spaces. Neither collenchyma nor stereome was observed; thus the mestome strands are located directly in the chlorenchyma; they are collateral and are surrounded by a green parenchyma sheath. The blade thus exhibits a very weak and simple structure.

The petiole is hairy at the base from pluricellular glandular hairs (fig. 6), but otherwise it is perfectly glabrous. It is triangular in cross-section. The cuticle is somewhat wrinkled, and the epidermis consists of large, thin-walled cells. A compact, chlorophyll-bearing parenchyma surrounds a single central mestome bundle, which is orbicular in transverse section and shows a very distinct, thin-walled endodermis.

The stem leaves are sessile, very narrow, and scabrous along the margins from unicellular, prickle-like, and curved projections of the epidermis. Glandular hairs (fig. 6) are frequent along the margins of the stipules. The epidermis of the upper face shows a very pronounced striation of the cuticle (fig. 4); the lateral cell walls are undulate on both faces, and stomata with one pair of subsidiary cells were observed in equal number on both faces of the blade. While the cuticular striations appear as radiating from the center of the epidermal cells on the upper face, the striae on the lower face are parallel with the longitudinal axis of the leaf. In transverse sections the outer cell wall of the epidermis is moderately thickened and shows the elevated cuticle very distinct on the upper face (fig. 3). Otherwise the structure is identical with that of the basal leaves and shows the absence of mechanical tissues; the chlorenchyma, however, is less developed, there being only one layer of palisades, and the pneumatic tissue is composed of very irregularly shaped cells with wider intercellular spaces and contains raphides.

The characteristics of *H. coerulea*, therefore, are the lack of col-

lenchyma and stereome in the organs examined; the glandular hairs; the striate cuticle; and the presence of stomata on both faces of the cauline leaves.

#### HOUSTONIA PURPUREA

The rhizome is very short and condensed, being merely represented by short internodes, the basal portion of aerial shoots. Several axillary buds occur on these subterranean stem portions, and some of these develop during the fall into small rosettes of leaves with distinct petioles and green, almost glabrous, elliptical blades. Secondary roots occur in pairs at the nodes, between the leaves, and in addition other roots are formed near the middle of the subterranean internodes.

**The roots.**—The secondary roots are very slender, sparingly branched, and not very hairy. Their structure agrees in all essential points with those of the preceding species, but they differ in showing a slight increase in thickness, due to the development of cambial strata in the stele itself, but without influencing the structure of the peripheral strata from endodermis to epidermis. It appears as if the secondary formations are confined to the leptome and hadrome, and that they are of short duration, the roots persisting only for one or two seasons. The capillary lateral roots are diarch, with two vessels in the center and two broad groups of leptome.

**The aerial stem.**—The basal internodes are quadrangular, with the angles produced into short and very narrow wings. The cuticle is wrinkled longitudinally and quite thick. Unicellular, obtuse, and somewhat bent hairs are frequent; they are thick-walled and show longitudinal (not spiral) striae of the cuticle. Viewed *en face* the cells of the epidermis are rectangular, and the stomata have one pair of subsidiary cells parallel with the stoma. The outer and inner wall of the epidermis is thickened, but not the radial. Inside the epidermis is a cortex with chlorophyll and raphides, but there is no collenchyma, not even in the wings. The strata of the cortex are arranged very regularly in rays toward the center, and the innermost layer is differentiated as a thin-walled endodermis, with the Casparyan spots plainly visible. Directly bordering on the endodermis is an almost continuous band of leptome and hadrome with very narrow medullary rays. The central pith is solid but thin-walled, and con-

tains raphides but no starch. This same structure is to be observed in the upper internodes except that the outline becomes more sharply four-winged as the cortical parenchyma in the wings becomes collenchymatic, and the hairs are more abundant.

The peduncle of the flower, however, is not quadrangular, but cylindric, and the hairs are reduced to mere papillae, which are exceedingly numerous. There is no stereome and no collenchyma; the cortical parenchyma is thin-walled, very compact, and the endodermis consists of much larger cells than observed in the internodes. The structure of the central cylinder is the same.

**The leaves.**—The over-wintering leaves are petiolate; the petiole is triangular in outline and covered by a wrinkled cuticle. The epidermis is moderately thickened on the ventral face, but thin-walled on the dorsal. A thin-walled chlorenchyma with very little chlorophyll and raphides surrounds three veins, the median of which is the largest; it is crescent-shaped and contains a collateral mestome strand surrounded by a thin-walled, colorless parenchyma sheath. The lateral veins are much smaller and round in transverse section. The blades of these basal leaves are sparingly hairy on the upper face from short, unicellular, obtuse hairs with very distinct longitudinal ridges of cuticle; but where the cuticle covers the ordinary cells of the epidermis (not the hairs) on the upper face we meet with a corresponding striation as observed in *H. coerulea* (fig. 9A). The lateral cell walls of the epidermis are undulate on both faces, especially on the lower, and it seems characteristic of this species (when compared with *H. coerulea*) that the lateral walls show local thickenings (figs. 9 and 9A). This structure I observed, also, and much farther developed in *Mitchella*, where it will be described more particularly. Stomata occur only on the lower face and show the same structure as those of *H. coerulea*. There is a typical palisade tissue of two layers, and a very open pneumatic tissue with the cells oblong and parallel with the leaf surface. The mestome strands are surrounded by parenchyma sheaths, but have no mechanical support, neither of collenchyma nor of stereome.

The stem leaves are sessile and their structure differs somewhat from that of the basal leaves. For instance, the cuticle does not show the stellate striations, and bicellular hairs (fig. 8) abound on

the upper face and along the margins, also along the veins on the lower face; moreover the lateral walls of the epidermis are straight, not undulate, on the upper face and do not show local thickenings. Cells with raphides are frequent in the pneumatic tissue, and a collenchymatic tissue occurs on the leptome side of the midrib. All the mestome bundles are collateral, and the median is the largest. Glandular hairs like those described and figured for *H. coerulea* were observed along the margins of the stipules.

The characteristics of this species are the local thickenings of the lateral cell walls of the epidermis of the leaves, and the presence of collenchyma, which seems to be absent in *H. coerulea*.

MITCHELLA REPENS (ANTHOSPERMEAe B. et H.)

In systematic works Mitchella is generally called an herb; "a smooth and trailing small evergreen herb" in the 6th edition of GRAY'S *Manual*, or "a small creeping evergreen" in GRAY'S *Synoptical Flora*. By NUTTALL it was considered as "an herbaceous repent evergreen;" BENTHAM and HOOKER describe it as "herba repens," with no allusion to the leaves being evergreen; while Linnaea in this same work (*Gen. Plant.*) is characterized as "fruticulus repens, sem-pervirens." Finally SCHUMANN (*Natürl. Pflanzenfam.*) describes Mitchella as "kriechende Kräuter" (*M. repens* and *M. undulata*); while in this same work *Linnaea borealis* is called "niederliegende Sträuchlein." Linnaea and Mitchella are both evergreen and both possess creeping, woody stems. MITCHELL, who was the first to describe our plant, called it Chamaedaphne, which shows that he had the correct impression of the plant as being an "undershrub" and not an "herb." Moreover, a plant cannot at the same time be an herb and an evergreen, and Mitchella is an undershrub in exactly the same sense as is Linnaea.

The stems are creeping, and the roots develop usually near the nodes and often two together, or commonly they develop from one to three at some distance from the nodes, and mostly from the lower face of the internode; the color of the roots is yellowish or light brown. All the internodes are stretched, and the opposite leaves have distinct petioles and green blades, the outline of which may vary from ovate to almost orbicular; no scale-like leaves develop in any place on the

floral or vegetative shoots. Very frequently some very long and purely vegetative shoots are to be observed in mature specimens, and from these develop lateral (axillary) floral shoots, which generally are quite short and bear only a few pairs of leaves, very often only two, though four or five are not uncommon. Whether the vegetative shoots stay as "vegetative," the ramification thus being monopodial, I am not in a position to state. As a matter of fact, it seems very difficult to make out the ramification of *Mitchella*, even where it occurs in abundance. The difficulty lies in the fact that the younger stages are hard to find and the adult specimens always appear as if they were not entirely complete. Injury to the shoots, for instance, might be the cause of arresting the buds in their farther and normal development.

The shoots of *Mitchella*, however, appear to be somewhat uniform, since their foliage is identical; and not as in *Linnaea*, where small, scale-like leaves occur, and the over-wintering buds are of two kinds. The ramification of *Linnaea* is doubtless much more complicated than that of *Mitchella*, but nevertheless it would be interesting to know exactly the behavior of the latter, how the shoots develop in young individuals, and especially how soon the stem becomes creeping.<sup>6</sup>

**The roots.**—The secondary roots are relatively short, but they ramify freely, and they remain active for several years. They are hairy and possess an exodermis of the same structure as epidermis. The cortical parenchyma is compact and consists of about three strata with the cell walls moderately thickened throughout; the endodermis, on the other hand, is thin-walled and small-celled; cells

<sup>6</sup> Professor WITROCK has presented a most excellent sketch of the biology of *Linnaea borealis* in *Botaniska Notiser* (1878-79), and calls attention to the fact that the earliest stages are yet unknown. In order to comprehend fully the morphology of *Linnaea* the following questions must be answered: (1) When does the main stem become decumbent? (2) Is the main stem a direct continuation of the primary axis, or is it developed from an axillary bud? (3) How soon does the ramification begin? (4) When does it commence to bloom? (5) Does the primary root stay unbranched, and how does it remain active? (6) When do the secondary roots develop? These questions may apply to *Mitchella* also, and I may at the same time quote another paper on *Linnaea* by Dr. SERNANDER (*Bot. Not.* 1891:225), in which these questions have been discussed and several additional observations recorded. (See also ARE-SCHOU *Bot. Not.* 1879:1.)

containing raphides occur in the cortex proper. The pericambium is thin-walled and continuous. Secondary formations take place at an early stage of these roots and thus the primary structure is obscured; the leptome occurs as several broad strands outside a much broader zone of hadrome, with many rows of vessels and thick-walled conjunctive tissue.

While studying the roots I noticed the peculiar fact that in some of these the lateral branches did not penetrate the cortex at once, but remained enclosed for some distance, thus traversing the cortical parenchyma although fully developed. The structure of these, however, was normal and they were developed from the pericambium. Such cases were merely exceptional, however, but may nevertheless deserve mention. Similar structures have been recorded for *Carpocephalus* and *Eriocaulon*.<sup>7</sup>

**The stem.**—An internode from a long, vegetative shoot of the previous year is cylindric, though somewhat flattened on account of its horizontal direction of growth, and it is very smooth. The cuticle is wrinkled, and the epidermis is quite thick-walled; hairs and stomata occur, but they are not frequent. Tannin was observed in the epidermis, also in the peripheral strata of the cortex, which is thin-walled and compact, contains raphides, and occupies the greater portion of the cross-section. A thin-walled endodermis, with the Casparyan spots plainly visible, surrounds the central cylinder. In the outermost stratum of this, thus bordering directly on the endodermis, I noticed about four very thick-walled cells, which resembled stereome but which proved to be secretory cells like those recorded by SOLEREDER as characteristic of *Mussaenda*, *Isertia*, *Cinchona*, and a few other genera; they are quite long and very thick-walled. The stele consists of many leptome strands and broad rows of vessels with narrow (one single row) medullary rays, while the center is occupied by a thick-walled, solid pith.

In shoots of this year's growth the structure is the same, but cells containing raphides are more abundant, and the stele shows its primary structure more distinctly, with the leptome and hadrome arranged in two large groups on the sides of the flattened internodes.

The erect shoots are strictly cylindric in outline, but show the

<sup>7</sup> HOLM, T., *Eriocaulon decangulare* L. BOT. GAZETTE 31:20. 1901.

same structure as the horizontal; however, they are more hairy and the cortex contains chlorophyll and raphides. In the first year of growth these shoots have no thick-walled secretory cells, and although the leptome constitutes an uninterrupted zone, the hadrome forms only two arches, separated from each other by two very broad rays of parenchyma. The stems of *Mitchella* increase very little in thickness, but they are woody and persist, as stated above, for several years.

The leaf-blade.—The cuticle is not very thick, but distinct; it is smooth on the lower face of the blade, except below the midrib, but wrinkled on the upper. The structure of the epidermis is somewhat peculiar. Viewed *en face* the lateral cell walls are very prominently undulate on both faces (*fig. 10*); besides that, local thickenings of these cell walls are very conspicuous. A transverse section (*fig. 11*) shows this structure perhaps more clearly, and the thickenings are seen to extend from the outer cell wall to the inner as columns. Whether this structure be common or not, it has not been recorded as characteristic of any of the Rubiaceae in the work of SOLEREDER. It was first described and figured apparently by MOHL<sup>8</sup> in the leaf of *Helleborus foetidus*, which also is an evergreen. However, it does not seem as if this particular structure is characteristic of plants with persisting leaves, since LALANNE,<sup>9</sup> who examined several genera of various families, does not mention any cases similar to the one described above. But in a paper by GODFRIN<sup>10</sup> a somewhat analogous structure is mentioned as occurring in the cotyledons of *Aesculus Hippocastanum* and *Acer platanoides*; and PÉE-LABY<sup>11</sup> records a similar case from the cotyledons of *Mimosa pudica*; the thickenings, however, do not extend to the bottom of the cell in these instances, but only to the middle.

The outer cell wall of the epidermis is moderately thickened on

<sup>8</sup> MOHL, HUGO VON, Grundzüge der Anatomie und Physiologie der vegetabilischen Zelle. Braunschweig. 1851. p. 14.

<sup>9</sup> LALANNE, GASTON, Recherches sur les caractères anatomiques des feuilles persistantes des Dicotylédones. Bordeaux. 1890.

<sup>10</sup> GODFRIN, J., Recherches sur l'anatomie comparée des cotylédons et de l'albumen. Ann. Sci. Nat. Bot. VI. 19:90. 1884.

<sup>11</sup> PÉE-LABY, E., Recherches sur l'anatomie comparée des cotylédons et des feuilles des Dicotylédones. Toulouse. 1892. p. 19.

both faces of the blade, and the lumen of the cells is much wider on the upper than on the lower face. The stomata (*figs. 12, 13*), which are confined to the lower face, are level with the epidermis; they have mostly two subsidiary cells parallel with the stoma and the air chamber is wide, but shallow. No hairs were observed. The chlorenchyma is differentiated into a palisade tissue of two strata and a pneumatic tissue of four layers with broad intercellular spaces (*fig. 14*). Cells with raphides were observed in the palisade tissue, whose cells diminish in size above the larger veins and become roundish. An almost colorless tissue surrounds the midrib completely and is collenchymatically thickened below this, but otherwise the veins have no mechanical support, since no stereome is developed. The mestome strands are collateral, and the median is very broad, with a large group of leptome and about three rows of vessels; a thin-walled parenchyma sheath surrounds each mestome bundle.

**The petiole.**—The cuticle is wrinkled and the outer walls of the epidermis moderately thickened. Very thick-walled bicellular hairs occur along the sides of the petiole, while at the base, and especially upon the inner face, numerous glandular hairs (*fig. 14, b*) were observed. Similar hairs were also found along the upper margins of the minute stipules. The chlorenchyma is quite open and contains only a little chlorophyll, but many raphides. A single, broad mestome bundle occupies the center; it is collateral and surrounded by a colorless, thin-walled sheath. The local thickenings of epidermis observed in the blade do not occur in the petiole.

The characteristics of *Mitchella*, therefore, are the structure of the epidermis of the leaf-blade; the long and slender glandular hairs; and the secretory, thick-walled cells inside the endodermis of the stem. The peculiarity noticed in some of the roots, where the lateral branches remain enclosed within the cortex for some time, is hardly to be considered as characteristic of the genus, since it was not found to be constant.

#### DIODIA TERES (SPERMACOCEAE B. et H.)

A seedling is shown in *fig. 15*. The primary root is quite long, sparingly hairy, and the lateral branches short. The hypocotyl is very distinct, erect and hairy, especially above, and the epigeic coty-

ledons are sessile (*fig. 16*) and hairy near the base on both faces; the stipules are represented by two linear, very small lobes, which bear very long and sharply pointed hairs. The leaves succeeding the cotyledons show the typical shape of the species and the stipules consist of several linear, hairy lobes. In mature specimens, which are annual, the primary root becomes very strong and woody; the main stem is erect, with many horizontal or ascending lateral branches, and the rigid, scabrous leaves are sessile and horizontal.

**The roots.**—The primary root shows secondary formations at an early stage, so that the primitive organization of the stele becomes obliterated. The epidermis, the cortex, and the endodermis become thrown off, and a cork of about four strata is developed from the pericambium; a large mass of leptome, hadrome, and thick-walled conjunctive tissue occupies the greater portion of the cross-section. In lateral roots of the first order epidermis is partly thrown off and there is no exodermis. The cortical parenchyma consists of four layers of large, thin-walled cells; the endodermis is also thin-walled and shows the Casparyan spots very plainly. Divisions have commenced to take place in the pericambium, and arches of cambium inside the leptome and outside the proto-hadrome have developed. The primitive structure is yet observable, there being five rays of hadrome alternating with five groups of leptome.

The lateral roots of the second order are capillary and show no increase in thickness. The epidermis is hairy and covers directly the cortex of two layers of very large, thin-walled cells. The endodermis is thin-walled, and the pericambium, which is continuous, surrounds two groups of leptome and one diametric row of narrow vessels.

**The stem.**—Numerous bicellular and rather thin-walled pointed hairs cover the stem from apex to base; the outline of the internodes is obtusely quadrangular, with two concave and two slightly convex faces. The cuticle is very distinctly wrinkled except over the hairs, where it is perfectly smooth. The epidermis is moderately thickened on the outer cell walls, and stomata are quite frequent. A few strata of collenchyma in a continuous zone separate the epidermis from the cortex, which consists of about four layers with narrow intercellular spaces. Chlorophyll was observed in the cortex, though in small

quantity. The innermost layer of the cortical parenchyma is differentiated as an endodermis (*fig. 21, end*), in which the inner and radial walls are very heavily thickened. The central cylinder contains a closed ring of leptome and about four rows of very narrow scalariform vessels and medullary rays of but one row between each two mestome strands. A thin-walled, solid pith occupies the center of the stem. Cells with raphides are frequent in the stem, but it seems as if they are confined to the cortex.

**The leaves.**—As mentioned above, the leaves are held in a horizontal position, but nevertheless their structure is almost isolateral, so far as the distribution of stomata and the differentiation of chlorenchyma are concerned. The cuticle is quite thick and smooth on both faces of the blade except the midrib, where it shows prominent longitudinal striations. Viewed *en face* the epidermis consists of pentagonal or hexagonal cells on the upper face, but of more irregular cells on the lower; the lateral cell walls are straight and not undulate in any part of the leaf. The stomata are most frequent on the dorsal face (*fig. 19*); the subsidiary cells, normally one pair, sometimes occur one on one side of the stoma and two on the other side, but all parallel with the stoma (*fig. 20*). Similar irregularities were also noticed in some of the other Rubiaceae examined, as may be seen from my figures. Epidermal projections abound in *Diodia*; the most common are small warts (*figs. 17 and 18*) developed merely as protuberances of the outer cell wall; they are very minute, but so numerous that they render the leaves scabrous. Unicellular, rather thin-walled, pointed hairs are also frequent (*fig. 19*) on both faces of the blade; and along the margins are curved, thick-walled, spine-like projections of exactly the same structure as those well known on the leaves of Gramineae and Cyperaceae (*fig. 18*).

A cross-section of the leaf shows a large-celled epidermis on both faces, with the outer wall slightly thickened on the upper; the warts appear most numerous on the upper face. The position of the stomata differs on the two surfaces, being raised on the lower and level with the epidermis on the upper. The midvein shows a prominent group of collenchyma and thin-walled water-storage tissue on the leptome side, while no collenchyma was observed above the hadrome. An isolated strand of collenchyma occurs also in the margins of the

leaf, but none accompanies the smaller veins. No stereome is present in the leaf. The chlorenchyma is quite compact throughout and is differentiated into a typical palisade tissue of two strata on the upper face of the blade and around the nerves. Between the nerves on the lower face of the blade the chlorenchyma consists of a pneumatic tissue of more or less oblong cells, with the intercellular spaces quite distinct but not so wide as in the other genera examined; raphides were observed in the pneumatic tissue, but not many among the palisades.

The mestome strands are collateral and surrounded by a very large celled and thin-walled parenchyma sheath, some of whose cells contain tannin; the midrib is the largest and its outline in cross-section is oval, while the others are much thinner and orbicular. The venation of the leaf is difficult to observe without separating the chlorenchyma from the veins, since the cross veins (secondaries) are so exceedingly thin and completely surrounded by chlorenchyma. The cross-veins are so numerous, indeed, that a transverse section of the blade shows their parenchyma sheaths so distinct "longitudinally" that the leaf is almost divided by them into two zones.

The stipules are divided into several bristles, which are terminated by a pointed hair, and a few (mostly three) glandular hairs occur at the base between the bristles; the structure of these glandular hairs is like those of *Houstonia* (fig. 6), but the stalk is much longer in *Diodia*. According to WARMING,<sup>12</sup> *D. radicans* Cham. et Schl. possesses dorsiventral leaves with the pneumatic tissue consisting of stellate cells with very wide intercellular spaces; moreover, the stomata, which are said to have no subsidiary cells, are confined to the dorsal face; hence the leaf is strictly dorsiventral.

*Diodia teres*, therefore, shows the following points of interest: the structure of the epidermis of the leaves; the approximately isolateral blade with stomata on both faces and with the palisade tissue partly extending to the dorsal epidermis; and the scant development of collenchyma and total absence of stereome in the leaves as well as in the internodes.

<sup>12</sup> Halofyt-Studier. Kgl. Danske Vid. Selsk. Skr. VI. 8:187. 1897.

## GALIUM (GALIEAE B. et H.)

WYDLER<sup>13</sup> has described the seedling and the structure of the shoots in several species of Galium. His paper contains many points of interest, for instance that the axes of first order in *G. cruciata* do not become terminated by inflorescences, and that accessory buds are frequent in European species. The seedlings of *G. saccharatum* and of *G. Aparine* are figured and described by LUBBOCK.<sup>14</sup>

## G. PILOSUM

Seedlings with the first two or three internodes developed may be found in the early part of May. The primary root (fig. 22, r) is quite long and branches freely, and is of a yellowish-brown color. The hypocotyl (fig. 22, h) is very distinct and bears epigeic, petiolate cotyledons with broad blades (fig. 23). While the hypocotyl is cylindric and glabrous, the succeeding internodes are quadrangular and hairy.

During the first season the hypocotyl becomes bent toward the surface of the ground and secondary roots commence to develop on all sides. The primary root continues to grow in length as well as in thickness. At the end of the first season the primary shoot above the cotyledons dies off, the hypocotyl thus being the only portion of the primary axis above ground that remains and winters over. At this stage, however, two minute buds become visible in the axils of the cotyledons, which in the next spring grow out into leafy shoots. Our species is a perennial herb, whose shoots die down to the ground every fall, while the buds in the axils of the basal leaves winter over, and the same mode of growth is repeated. I have never observed more than one bud in each axil, this species differing in this respect from those studied by WYDLER.

In full-grown, fruiting specimens of *G. pilosum* the primary root persists as a slender, woody taproot of a yellowish-brown color, which changes to a brilliant red when preserved in alcohol. There are many quite strong lateral roots and secondary ones, which develop very freely from the basal internodes of the stem. No real rhizome becomes developed; the persisting stem bases with their buds and

<sup>13</sup> Kleinere Beiträge zur Kenntniss einheimischer Gewächse. Flora 43:492. 1860.

<sup>14</sup> Seedlings 2:59. 1892.

the primary root are the only underground organs that winter over, and by which the future development of aerial floral shoots is secured. The term "pseudo-rhizome" seems very applicable to the subterranean stem of this and other species of the genus. It was originally suggested by HJALMAR NILSSON<sup>15</sup> and intended for herbs "in which the basal, subterranean portion does not die off with the aerial, but stays active and produces a floral shoot the succeeding year by means of a lateral bud; and in which this basal portion becomes able to increase in thickness and to nourish the over-wintering bud, and also to develop secondary roots; such a subterranean stem represents a pseudo-rhizome."

The roots.—The primary root persists for several years, and evidently as long as the individual lives. During the first season secondary formations in the stele commence, but the growth is rather slow and causes neither the cortex nor the epidermis to be thrown off. The epidermis thus remains intact, is slightly thickened on the outer cell walls, and is almost glabrous. There is no exodermis, and the cortex represents a compact tissue of about five strata in which cells with raphides are scattered. The endodermis is thin-walled and readily distinguished by the small lumen of the cells when compared with those of the adjoining cortex. The pericambium is also thin-walled and continuous and does not yet (August) show any cell division; but there are additional vessels inside the leptome, so that the primitive structure of the stele is no longer to be observed; the medullary rays are very narrow, consisting only of one row of cells.

The lateral roots of the first order are almost capillary, and the thin-walled epidermis shows many hairs. The cortical parenchyma is very thin-walled and the cells large. A thin-walled endodermis and pericambium surround two groups of leptome and two very narrow vessels in one diametrical plane.

While studying the structure of the primary root of a number of specimens I noticed the same peculiarity as in *Mitchella*, that the lateral roots stay for some time enclosed within the cortex, which they traverse longitudinally; this peculiarity was noticed in several cases, but not frequently enough to make me believe that it is characteristic of this particular species.

<sup>15</sup> Dikotyla jordstamar. Acta Univ. Lund 19:18. 1882-83.

**The stem.**—The basal internodes are four-winged, and very hairy from long, unicellular hairs with the cuticle minutely granular (*fig. 24*); otherwise the cuticle is thick and perfectly smooth. A very pronounced thickening with layerings is observable in the outer cell wall of epidermis, the stomata are raised a little above the adjoining epidermis, and the air chamber is deep but narrow. A broad strand of about six layers of very thick-walled collenchyma is located in each of the four wings, just beneath epidermis. Viewed in longitudinal sections the cells of this collenchyma showed the cross walls horizontal to very oblique. The cortex consists of about seven strata inside the wings and of only three between them; it is thin-walled and quite open on account of the very wide intercellular spaces; cells with raphides are frequent in this parenchyma, but no starch was observed. The innermost layer is differentiated as a thin-walled endodermis, directly bordering on a continuous zone of leptome, inside of which is a very broad zone of hadrome with the vessels narrow and thick-walled. The center of the stem is occupied by a thin-walled pith, mostly broken down in the middle. This same structure was observed also in the internodes of the mature stems, but not in the lateral axes that bear the inflorescences. In these the wings are less distinct and the pith is solid. Finally, in the floral peduncle the outline is cylindric and the collenchyma totally absent.

**The leaves.**—An isolateral structure is characteristic of the cotyledons. The lateral cell walls of the epidermis are undulate on both faces, and stomata occur on the dorsal as well as on the ventral face. The chlorenchyma is represented by a homogeneous tissue throughout, of cells more or less oblong to roundish in transverse section, and no palisades were observed. Several of the epidermal cells contained brown, amorphous clumps of resin, and these secretory cells in the cotyledons did not differ from the other cells of epidermis. The slender petiole has only one broad mestome bundle in the middle and no mechanical tissue.

The small stem leaves and the foliaceous stipules show the following structure. The cuticle is smooth except above the large epidermal cells which contain resin, where it is irregularly thickened so as to form striations (*fig. 25*). Viewed *en face* the lateral walls of the

epidermis are undulate on both faces, and many very large secretory cells occur on the dorsal, besides stomata; the latter have mostly one pair of subsidiary cells parallel with the stoma; unicellular hairs, often with apex hooked, abound on the dorsal face. In transverse sections the epidermis appears to be slightly thickened on both faces, and the lumen of the cells is somewhat wider on the upper than on the lower, excepting the secretory cells, which, as stated above, are very large. The stomata are raised, and in the leaves the air chamber seems constantly to be wide but shallow.

A chlorenchyma is represented by one layer of palisades on the upper face and by three or four strata of very open pneumatic tissue on the lower. Three thick-walled layers of collenchyma are located on the leptome side of the midrib, inside the epidermis; no stereome was observed. A water-storage tissue of thin-walled, colorless cells in about five layers in a very broad group cover the leptome, making the midrib very prominent on the dorsal face. Neither the collenchyma nor the water-storage tissue was observed at the other nerves. Cells containing raphides were only found in the chlorenchyma, between the palisades and the pneumatic tissue. All the nerves are surrounded by parenchyma sheaths of small, rectangular cells with the Casparyan spots very plainly visible, having more the aspect of an endodermis than of the usual parenchyma sheath in leaves. The midrib is the thickest on account of the very broad collateral mestome strand, and because accompanied by collenchyma and water-storage tissue. The lateral veins are very thin, and orbicular in cross-section; they are collateral and the parenchyma sheath is very distinct as in the midvein.

The characteristics of *G. pilosum* are the leaf epidermis with resiniferous cells and their cuticular striations. Secretory cells in epidermis are recorded by SOLEREDEER (p. 505) from Rubia, Anthospermum, and Nenax, but not from Galium. They are mentioned however, by KEARNEY, who found them in the leaves of *G. hispidulum* Michx.<sup>16</sup> The collenchyma is well represented in the stem, but is confined to the midrib in the leaves. The coloring matter observed in the root is well known in other species of Galium, as well as in Rubia.

<sup>16</sup> Report on a botanical survey of the Dismal Swamp region. Contrib. U. S. Nat. Herb. 5:506. 1901.

**G. TRIFLORUM**

The seeds of this species germinate very early, and seedlings may be found in wooded ravines in February. The hypocotyl (*fig. 28, h*) is very long and lies on the ground; the primary root is distinct and amply branched. Very characteristic are the large blades of the cotyledons and the relatively short petioles. Small shoots are already developed in the axils of these, while the primary axis, which is erect, shows long, glabrous internodes. During the first season the plant thus develops three aerial shoots, which however die down to the ground in the fall. The hypocotyl and the primary root do not persist, but the base of the two axillary shoots from the cotyledons remains alive, and the future growth of the individual is secured by small buds in the axils of the basal leaves and some secondary roots that winter over. In mature specimens the root system is thus represented by secondary roots, and the subterranean stem is a pseudorhizome like that of the preceding species.

**The roots.**—The secondary roots are very long and relatively thick and strong; they become glabrous very soon. No exodermis is developed, and the cortex, which is very compact, consists of four starch-bearing layers. The endodermis and the continuous pericambium are thin-walled, and the stele shows three leptome strands alternating with three rays of hadrome; while the center is occupied by a thick-walled conjunctive tissue. It appears as if the increase in thickness does not commence until during the winter, when the epidermis and the cortex become thrown off.

The lateral roots are capillary and almost destitute of hairs; there is no exodermis and the structure is identical with that of the mother root, but they do not increase in thickness. I observed no case where these lateral roots remained enclosed within the cortex, as in the former species.

**The stem.**—The internodes are quadrangular and sharply four-winged; unicellular, pointed hairs are common along the wings, rendering them quite scabrous. The thin cuticle shows longitudinal ridges, and the outer cell wall of the epidermis is slightly thickened. Broad groups of thick-walled collenchyma are noticeable in the wings, but otherwise the stem has no mechanical support. The cortex consists of about ten layers in the wings, and of three between them;

it contains chlorophyll and raphides. A thin-walled endodermis surrounds the central cylinder, consisting of a continuous zone of leptome, inside of which the vessels are arranged in short rows with narrow medullary rays bordering on the central, very thin-walled pith.

**The leaves.**—The structure is almost isolateral in respect to the chlorenchyma. The cuticle is thin and smooth on both faces, and the lateral cell walls of the epidermis show a very pronounced undulation (*fig. 26*); the outer cell walls are moderately thickened (*fig. 27*). Unicellular hairs and curved, prickle-like projections are common along the margins and the veins on the lower face, but no resiniferous cells were observed in this species. The stomata are slightly raised and have one pair of subsidiary cells with the lateral walls undulate. No palisade tissue is developed, the chlorenchyma being a homogeneous tissue of roundish cells (*fig. 30*) with distinct intercellular spaces, especially near the dorsal epidermis, where cells containing raphides are abundant. A small group of collenchyma and a water-storage tissue of a few cells cover the leptome side of the midvein. The mestome bundles show an orbicular cross-section, and are collateral; the parenchyma sheaths show the same structure as observed in the preceding species, with the Casparyan spots very plainly visible.

The structure of *G. triflorum* thus resembles that of *G. pilosum*, but is generally weaker, the epidermis does not show the resinous secretions, and the leaves are almost isolateral.

#### G. CIRCAEZANS

I have not been able to find the seedlings of this species, and must therefore confine myself to the mature plant. It is very distinct from the two species described above in the relatively long and broad leaves and stipules, also in possessing a true rhizome. The stem bases persist and buds become developed upon them, some of which grow out directly as aerial shoots, while others stay underground, producing stolons with stretched internodes and scale-like leaves. The primary root does not persist, but becomes replaced by numerous secondary roots from the nodes and internodes.

**The roots.**—The secondary roots are long, slender, and profusely branched; light brown when fresh, but placed in alcohol they attain

rapidly a beautiful crimson color. The epidermis is glabrous and very small-celled; it covers the cortex directly, there being no exodermis. The cortex is thin-walled, but compact; no starch was observed, but globules of resin occur in some of the cells; it consists of about six strata, the innermost of which is differentiated as a small-celled, thin-walled endodermis. Secondary formations have changed the primitive structure of the stele, which shows a confluent zone of leptome and many rows of vessels separated by a few strata of thick-walled conjunctive tissue, which extend to the center.

The lateral roots are also glabrous and have no exodermis. About four layers of cortex with raphides border on a thin-walled endodermis. The pericambium is continuous and surrounds four broad groups of leptome alternating with four short rays of hadrome, with a central group of thick-walled conjunctive tissue. A lateral root of the second order (borne upon the former) is exceedingly thin and slightly hairy. In this the cortex consists only of two layers, the innermost of which is large-celled, with the cells stretched radially and containing raphides. The stele is diarch.

**The stem.**—The structure of the stolons is rather weak; the epidermis and the primary cortex had been thrown off, and the central cylinder was merely covered by some layers of cork. This cork was evidently developed from endodermis, as in other species of the genus and of *Rubia*, as stated by SOLEREDER (p. 510). The stele is located directly inside the cork and consists of a confluent zone of leptome separated from the hadrome by strata of cambium. A small pith occupies the center of the internode.

The aerial stem is sharply four-winged, covered by a thin cuticle, which is somewhat wrinkled along the wings, but otherwise smooth. The epidermis is glabrous and the outer cell wall thickened, especially in the wings. Several strata of heavily thickened collenchyma were observed beneath epidermis in the wings (*fig. 34, coll.*). The cortical parenchyma in the wings consists of about seven compact layers with chlorophyll, and of only three between them. The endodermis is thin-walled and surrounds the stele, which shows the same structure as in the preceding species.

**The leaves.**—The lateral cell walls of the epidermis are prominently undulate on both faces, when viewed *en face*. Cells with resin are

frequent, but only on the lower face (*fig. 31*). The cuticle is wrinkled on the upper face, but smooth on the lower, except over the hairs, where it is minutely granular. The epidermis is thin-walled; unicellular, seldom bicellular, hairs cover the lower face, especially the nerves; they are quite thick-walled and the apex is pointed and more or less curved. The stomata, which are confined to the lower face, have two subsidiary cells, one of which is mostly much smaller than the other, but they are both parallel with the stoma. A cross-section of the leaf (*fig. 32*) shows the lumen of the epidermis to be about the same on both faces; also the structure of the chlorenchyma, which shows a palisade tissue quite well distinguished from the underlying, very open, pneumatic tissue (*fig. 33*). A small group of collenchyma and water-storage tissue is located underneath the median and the two lateral mestome bundles. The structure of the veins agrees in all respects with that observed in the former species. Besides the secretory cells in the epidermis, there are in the pneumatic tissue many cells containing raphides.

In *G. circaeans* the collenchyma reaches a high development in the stem, but not in the leaf. The epidermal secretory cells show the same structure as those of *G. pilosum*, but the cuticle is uniformly smooth over them, not striate as in *G. pilosum*. The leaf structure is bifacial, even if the palisades are not quite typical.

#### G. LATIFOLIUM

In habitus this species resembles very much the preceding, but is more robust, taller, with coarse stems and larger leaves. The primary root does not persist, but secondary ones are developed in great numbers; they are yellowish-brown, quite thick, and branch freely. This species has no true rhizome, but a pseudo-rhizome of densely matted basal internodes, that persist for several years. The foliage of these over-wintering stem portions is reduced to small, scale-like leaves of the same length as the internodes. The aerial shoot is stout, with several lateral branches, all of which are terminated by inflorescences.

**The roots.**—The secondary roots are glabrous, and the epidermis is thin-walled. A compact cortex of three strata borders on the endodermis, of which the cell walls are moderately thickened; the peri-

cambium is thin-walled and continuous, with beginning cell division. Secondary formations have already commenced in the stele, where the primitive structure is no longer observable. A thick-walled conjunctive tissue occupies the greater portion of the central cylinder. Some of these secondary roots were quite thick, and were evidently two years old. The cortical parenchyma was thick-walled and consisted of ten compact strata, surrounding a very broad stele, in which the hadromatic rays were considerably longer than in the other roots.

The capillary, lateral roots are also without hairs, and they show the same structure as the mother root, the only differences being that the cortex is smaller and that they do not increase in thickness.

**The stem.**—The internodes above ground show the same structure as *G. circaeans*, but the wings are not so sharp (fig. 35); the collenchyma thus represents a broader group. In the basal internodes that winter over, the collenchyma is much reduced (fig. 36), while the cortex is frequently more or less thick-walled.

**The leaves.**—There is only one point in regard to the internal leaf-structure in which this species differs from *G. circaeans*, namely the resiniferous cells in the epidermis. These are much more numerous and often occur several together, and viewed *en face* they show branches very often, so as to acquire a very irregular outline, instead of being merely oblong. Over these cells the cuticle is prominently striate, as observed in *G. pilosum*. The only characteristic feature by which this species may be distinguished from the others, therefore, depends upon the abundance and much larger lumen of the secretory cells in the leaves.

#### SUMMARY

According to VESQUE,<sup>17</sup> the characteristics of the Rubiaceae are the simple hairs; the absence of glandular hairs; the stomata accompanied by at least one pair of subsidiary cells parallel with the stoma;<sup>18</sup> the crystals simple, druids, raphides, or as crystalline sand; the absence of laticiferous ducts. The peculiar glandular hairs which I found in *Mitchella* and *Houstonia* are not glandular hairs in the

<sup>17</sup> Caractères des principales familles gamopétales tirés de l'anatomie de la feuille. Ann. Sci. Nat. Bot. VII. 1:192. 1885.

<sup>18</sup> In accordance with WARMING the stomata of *Diodia radicans* Cham. et Schl., from the sandy shore of St. Thomas, have no subsidiary cells. Halofyt-Studier, p. 187.

same sense as this term is generally used. SOLEREDER (p. 504), who records them from the stipules of *Isetia* for instance, calls them *Drüsenzotten*, but I am at present unable to suggest any better English name than "glandular hairs." The other characters enumerated by VESQUE are readily recognized in the plants described above.

The epharmonic variations are also discussed by VESQUE (p. 202), but these are not exactly comparable with those observed in our plants, since the genera treated by VESQUE are so very different. However, the same degree of variation takes place in several instances, and we might consider these epharmonic characters as suggested by VESQUE. Among the epharmonic variations observed in the Rubiaceae described above, the following may be enumerated:

**The roots.**—The superficial development of cork inside the exodermis in *Cephalanthus*; the lack of exodermis in *Houstonia*, *Diodia*, *Oldenlandia*, and *Galium*; the thick-walled cortical parenchyma in *Mitchella* and *G. latifolium*.

**The stem.**—The presence of stereome in *Cephalanthus*; the development of collenchyma as a continuous zone in the same genus and in *Diodia*; the isolated collenchyma strands in the angles of the stem in *Houstonia purpurea* and *Galium*; the lack of collenchyma in *Oldenlandia*, *H. coerulea*, and *Mitchella*.

**The leaves.**—The bifacial structure in *Cephalanthus*, *Oldenlandia*, *Houstonia*, *Mitchella*, *Galium circaeans*, *G. latifolium*, and *G. pilosum*; the isolateral structure in *Diodia* and *G. triflorum*; the distribution of the stomata on both faces of the (cauline) leaf-blade in *H. coerulea* and *Diodia*; the presence of epidermal resin cells in *G. pilosum*, *G. circaeans*, and *G. latifolium*; the local thickenings of the lateral cell walls of the epidermis in *Mitchella* and *H. purpurea*; the straight rather than undulate lateral cell walls of the epidermis in *Cephalanthus*, *H. purpurea* (the cauline on the upper face), and *Diodia*; the cuticular striae radiating from the center of the epidermal cell in *H. coerulea* and *H. purpurea*; the cuticular spiral striations over the hairs in *H. coerulea*; the wrinkled cuticle above the resin cells in *G. pilosum* and *G. latifolium*; the granular cuticle over the hairs in *G. pilosum*, *G. circaeans*, and *G. latifolium*; the glandular hairs in *Oldenlandia*, *Houstonia*, and *Mitchella*; the palisade tissue extending to the dorsal epidermis in *Diodia*; the non-development of typical palisades

in *G. triflorum*; the lack of collenchyma in *H. coerulea* and in the over-wintering leaves of *H. purpurea*; the presence of a water-storage tissue, amply represented in *Cephalanthus*, and only on the leptome side of the midrib and larger veins in *Galium* and *Diodia*; the endodermis-like parenchyma sheath in *Galium*.

Some of these characters may not be constant. For instance, the relative development of the palisade tissue may be different, and it would be interesting to know the leaf-structure of *Galium triflorum* from northern Europe, and of the Japanese *Mitchella*.

The family characters, as outlined by VESQUE and SOLEREDER, may be augmented, but hardly changed. The family is well represented in North America, and an extended study of the southern and western species would no doubt give many interesting results.

BROOKLAND, D. C.

#### EXPLANATION OF PLATES VII-IX

##### PLATE VII

###### *Cephalanthus occidentalis*

FIG. 1. Transverse section of root, showing development of cork; *ep*, epidermis; *ex*, exodermis; *p*, phellogen; *c*, outermost stratum of cortex.  $\times 320$ .

FIG. 2. Epidermis of lower face of leaf with stomata.  $\times 240$ .

###### *Houstonia coerulea*

FIG. 3. Cross-section of stem leaf, showing a cell of epidermis with its cuticle from the upper face.  $\times 240$ .

FIG. 4. Epidermis from upper face of stem leaf with cuticular striations and stomata.  $\times 240$ .

FIG. 5. Hair from leaf margin with spiral striations of cuticle.  $\times 240$ .

FIG. 6. Glandular hair from stipule.  $\times 240$ .

FIG. 7. Epidermis with stomata from lower face of basal leaf.  $\times 320$ .

###### *Houstonia purpurea*

FIG. 8. Hair from upper face of stem leaf.  $\times 240$ .

FIG. 9. Epidermis with stomata from lower face of basal leaf.  $\times 240$ .

FIG. 9A. Epidermis of upper face of basal leaf, showing cuticular striations and thickenings of the lateral cell walls.  $\times 320$ .

###### *Mitchella repens*

FIG. 10. Cells of epidermis from upper face of leaf, viewed *en face*.  $\times 560$ .

FIG. 11. Cross-section of leaf, showing epidermis (*ep*) and part of palisade tissue (*p*).  $\times 560$ .

FIG. 12. Epidermis of lower face of leaf, showing the stomata.  $\times 320$ .

FIG. 13. Epidermis of lower face of leaf.  $\times 320$ .

FIG. 14. Pneumatic tissue of leaf, viewed *en face*.  $\times 240$ .

FIG. 14B. Glandular hair from base of petiole.  $\times 240$ .

#### PLATE VIII

##### *Diodia teres*

FIG. 15. A seedling; *cot*, cotyledons; *h*, hypocotyl; *r*, primary root.  $\times 2$ .

FIG. 16. A cotyledon with stipule.

FIG. 17. Transverse section of leaf, showing epidermis of upper face with protuberances of outer cell wall.  $\times 240$ .

FIG. 18. Epidermis of leaf margin.  $\times 240$ .

FIG. 19. Epidermis of lower face of leaf.  $\times 240$ .

FIG. 20. Epidermis of upper face of leaf, showing stomata.  $\times 240$ .

FIG. 21. Cross-section of stem internode, showing endodermis (*end*) and cortex (*c*).  $\times 240$ .

##### *Galium pilosum*

FIG. 22. A seedling; index letters as in fig. 15.  $\times 2$ .

FIG. 23. A cotyledon.

FIG. 24. Hair from stem.  $\times 240$ .

FIG. 25. Epidermis of lower face of leaf, showing stomata and a secretory cell with cuticular striations.  $\times 240$ .

##### *G. triflorum*

FIG. 26. Epidermis of lower face of leaf, showing stomata and undulate lateral cell walls.  $\times 320$ .

FIG. 27. Transverse section of leaf, showing epidermis and a stoma from lower face of blade.  $\times 320$ .

#### PLATE IX

FIG. 28. A seedling; index letters as in fig. 15.  $\times 2$ .

FIG. 29. Hair from midrib.  $\times 240$ .

FIG. 30. Transverse section of ventral portion of leaf; *ep*, epidermis; *m*, mesophyll without palisade.  $\times 320$ .

##### *G. circaeans*

FIG. 31. Epidermis from lower face of leaf, showing a secretory cell with no cuticular striae.  $\times 240$ .

FIG. 32. Cross-section of leaf; *ep*, epidermis; *p*, palisade tissue.  $\times 320$ .

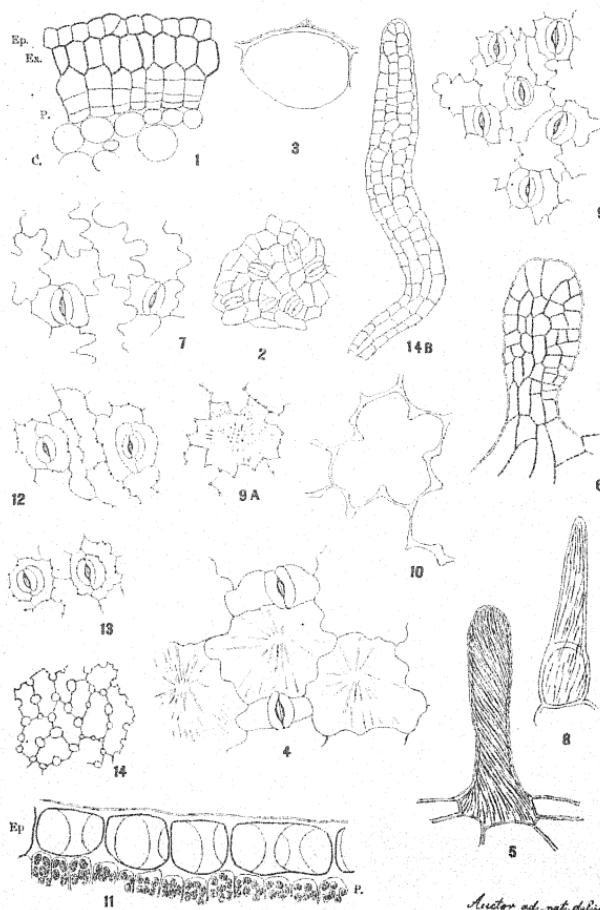
FIG. 33. Pneumatic tissue viewed *en face*.  $\times 320$ .

FIG. 34. Cross-section of internode, showing one wing; *ep*, epidermis; *coll*, collenchyma; *c*, cortex.  $\times 320$ .

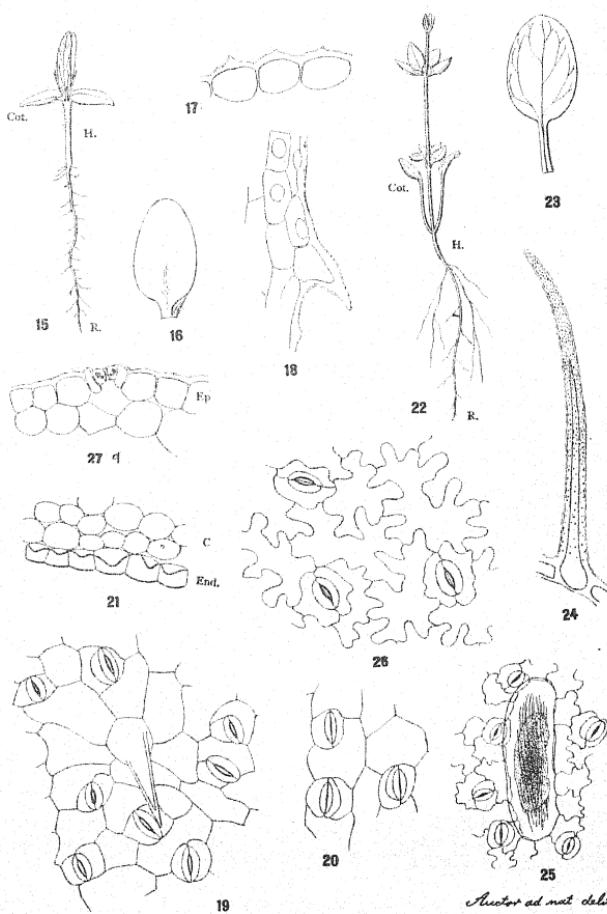
##### *G. latifolium*

FIG. 35. Cross-section of basal (but aerial) internode, showing one of the angles; index letters as above.  $\times 320$ .

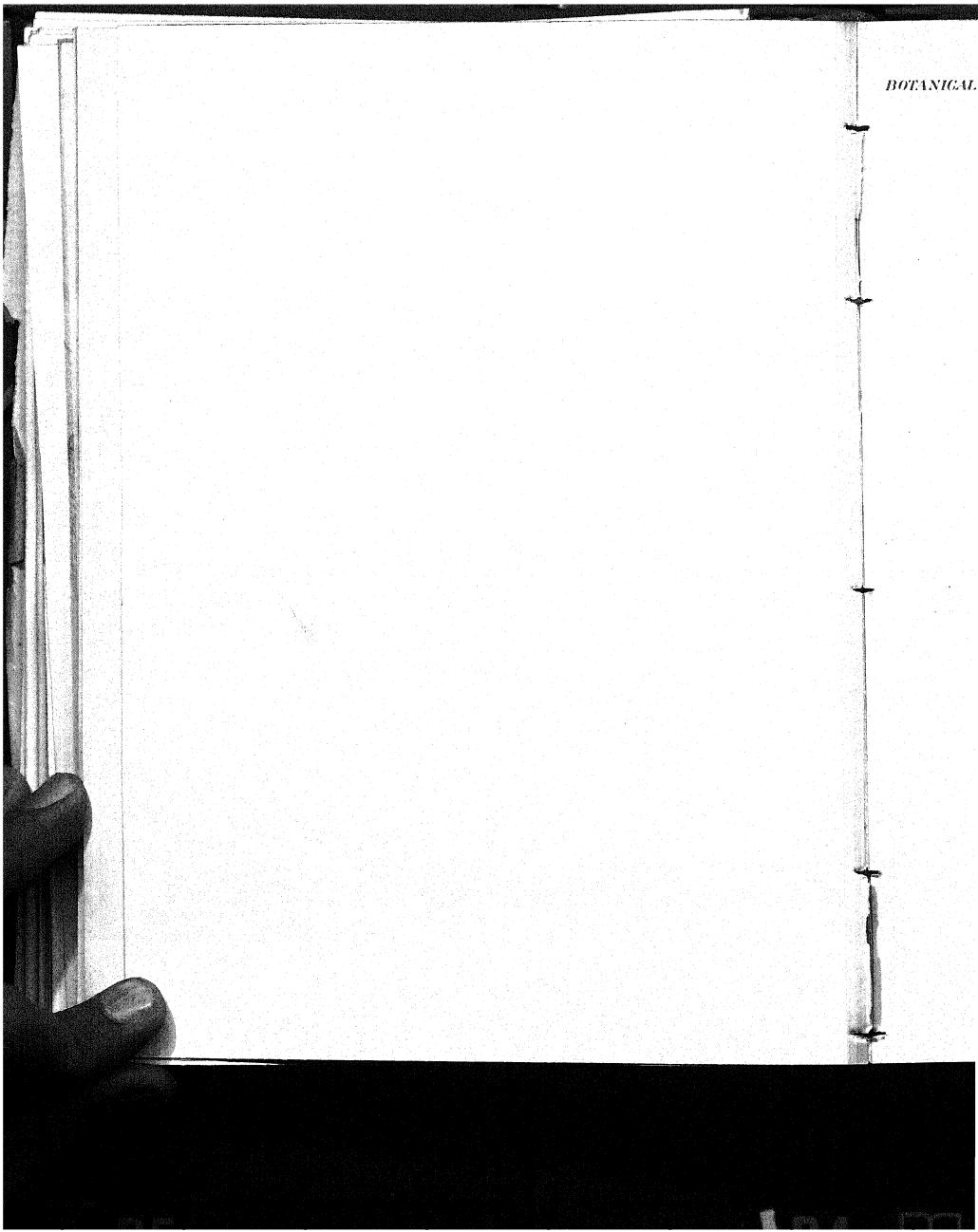
FIG. 36. Cross-section of subterranean internode of an over-wintering shoot; index letters as above.  $\times 320$ .

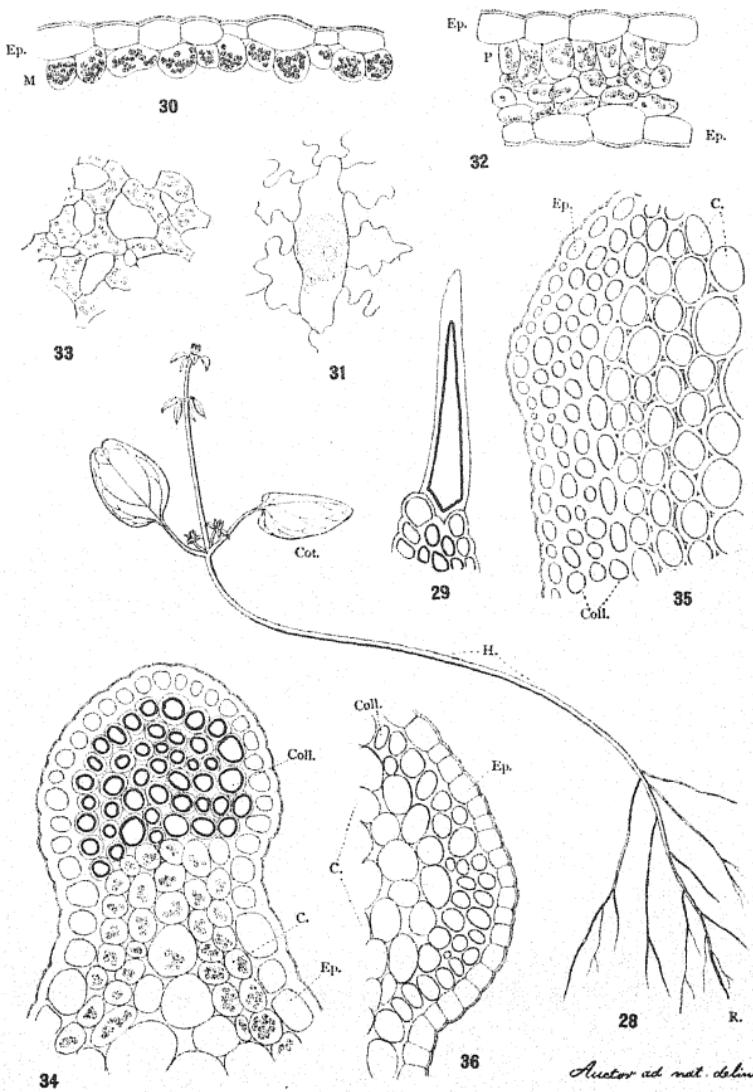
*Anator ad. nat. delin.*

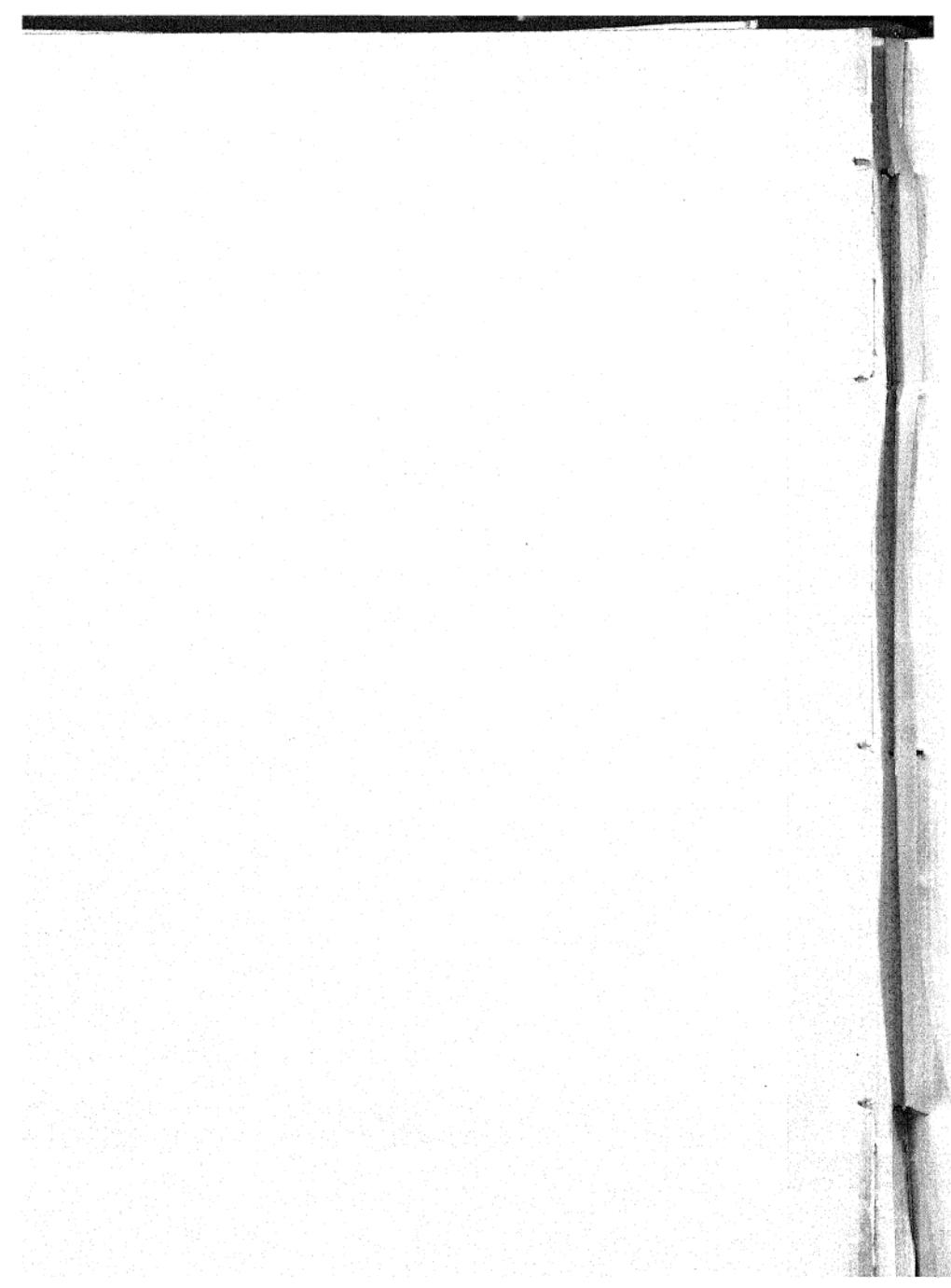




*Microrad not delin.*







MORPHOLOGY OF THE TRUNK AND DEVELOPMENT  
OF THE MICROSPORANGIUM OF CYCADS  
CONTRIBUTIONS FROM THE HULL BOTANICAL LABORATORY  
XCI

FRANCES GRACE SMITH

(WITH PLATE X)

The original purpose of the investigation recorded in this paper was to secure as complete an account of the development of the microsporangium of cycads as possible, and especially to discover whether the archesporium is several-celled, as indicated by previous investigations. In securing staminate strobili at the earliest stage, attention was drawn to their successive appearance and the building up of the trunk, and the results of these observations have been included.

The staminate strobili of *Ceratozamia mexicana* Brogn. (*C. robusta* Miq.) were obtained from the Botanical Gardens of Jena and Berlin, and also from Fairmount Park, Philadelphia. A strobilus of *Encephalartos villosus* was obtained from the Botanical Garden at Bonn; and most of the strobili of *Zamia floridana* were sent from Miami, Florida, in 1902, 1905, and 1906. No material of *Ceratozamia* was younger than the division of sporogenous cells just preceding the mother cells; and the material of *Encephalartos* showed sporangia about to shed the pollen. In *Zamia* the series is much more complete than in the other forms. Collections of staminate cones were sent from Miami every two or three weeks, from June 1 to September 18, 1905; then less frequently up to the last of January, 1906, when pollination took place. In 1906 further collections of staminate and ovulate plants were made, at intervals of ten days, from June 5 to September 1. These gave stages in the development of the strobili as well as of the sporangia. Only in *Zamia floridana*, therefore, has there been any attempt to study the early development of the sporangium or strobilus. The other genera have been used for comparison in the later stages of sporangium-development.

### I. Morphology of the trunk

The cycads have a columnar or tuberous trunk, thickly covered with scale leaves and old leaf bases. *Zamia floridana* illustrates a

somewhat intermediate type, in which the trunk is a little elongated, but still tuberous, the greater part of it being subterranean. The strobili of cycads apparently terminate the axis, and this position is so distinct from the laterally borne strobili of other gymnosperms that it assumes an importance demanding more than the brief statements given to it in the literature, all of which seem to be based upon observations of mature strobili.

Elongation of the stem by continued growth of the apical meristem is shown by all cycads until they begin to bear strobili, and continues to be the method of elongation of the ovulate plants of *Cycas*. In the staminate plants of *Cycas*, and in both staminate and ovulate plants of the other genera, however, a very different method of elongation sets in. This is evident from the fact that cycads live for many years after fruiting and produce a succession of crowns, so that the growth of the axis has not been stopped; and also from the fact that frequently more than one strobilus is produced at the summit of the stem, as in both ovulate and staminate plants of *Zamia*. The following brief historical sketch will serve to outline the views that have been held in reference to the relation of the strobili to the stem axis.

MIQUEL (1) does not commit himself to any theory as to the mode of branching. He finds the cones are elongated axes of the stem in *Cycas*, *Macrozamia*, and *Encephalartos*, but "perchance lateral axes, as in *Zamia*."

KARSTEN (2) states that "the flowers appear in *Zamia muricata* on the same stem, one after another," implying sympodial branching.

DEBARY (3) gives an instance of monopodial branching in connection with a staminate strobilus of *Cycas Rumphii*. At first the strobilus occupied a terminal position, but a vegetative point was found at its base. Later, when the strobilus had matured and had been cut off, the vegetative point assumed the terminal position; but because the strobilus showed a depression upon the side toward the vegetative point and the leaves of this point were stunted, DEBARY inferred that the staminate strobilus was really axillary, and that the vegetative point had been stunted and forced out of position. He gives the other possibility, namely, that the strobilus is really a terminal structure and is pushed to one side by the vegetative point; but he thinks that the facts stated above do not carry out this assumption.

tion, although he says that it must be proved in either case by stages from young cones.

BRAUN (5) definitely suggests sympodial branching, regarding the staminate and ovulate strobili as terminal, and quoting KARSTEN as given above. ENGLER and PRANTL (14) in turn quote BRAUN.

WARMING (7), in his investigation of Ceratozamia, decides that the staminate strobilus is terminal, and in an excellent drawing of a transverse section through leaves and strobilus shows the bud appearing in the axil of one of the leaves and continuing the principal axis as a sympodium. The peduncle of the strobilus is indicated between two leaves.

SACHS (9) and GOEBEL (11) state in effect that if the strobilus is single it stands as a terminus of the stem; but if there are two or more strobili, as in *Zamia muricata* and *Macrozamia spiralis*, they may be the result of the forking of the stem.

COULTER and CHAMBERLAIN (19) state that the staminate strobilus in Cycas, at least, is terminal; but that in the other genera its true position, although apparently terminal, remains in doubt. Strobili may occur singly or several together, and in the latter case, at least, are considered lateral.

WORSDELL (25) follows the theory of the sympodium in part, stating that each strobilus is developed as a continuation of the main axis, and that growth in length is continued by means of a lateral vegetative shoot immediately below the strobilus, the latter being pushed to one side. He also suggests monopodial branching, where, "as is frequently the case, short lateral shoots are developed at intervals upon the main axis."

This historical sketch shows that the branching of the cycadean trunk has been regarded by various writers as dichotomous, as monopodial, and as sympodial.

In preparing the material so that the strobili could be severed from the stem for killing, the petioles of old leaves and the scale leaves had to be removed carefully from the top of the stem; for often in the younger stages the strobilus is almost concealed by the growing tissue around it, only the tip projecting above the firm tissue of the stem. This tip is completely covered by scale leaves, so that often the strobili reach the stage in fig. 14 before the tip emerges from the scale leaves. In many cases more than one strobilus was.

found at the summit of the stem; and in connection with the young strobili the petioles of old leaves and peduncles of old strobili were *in situ*, so that the relative positions of the organs could be ascertained.

A stem received July 5 showed the remnant of an old peduncle, a young ovulate strobilus, and about 5<sup>mm</sup> from this a circle of leaves surrounding a growing point (*fig. 1*). Another ovulate plant of the same age had a young strobilus, but apparently no vegetative point; however, on sectioning this young strobilus and the tissue at its base, the vegetative point was disclosed very close to the strobilus, with a few leaves surrounding it. This growing point with its leaves was enclosed within the scale leaves, which in turn surrounded the strobilus (*fig. 2*).

Two other ovulate plants when sectioned showed each a smaller strobilus at the base of the larger one, and between them the vegetative point like a small protuberance surrounded by rudimentary leaves. This whole group was enclosed within a circle of leaves which formed a hollow cone (*fig. 3*).

One of the smallest staminate cones, received June 14, had a vegetative point at its base, with rudimentary leaves (*fig. 4*). In material received July 5 three staminate cones were found upon one plant (*fig. 5*). These were of different sizes and the vegetative point was situated at the base of the smallest cone. Each strobilus of the group had apparently been the terminal one, and had been pushed aside by the newer one in its development. The strobili would thus be in lateral positions at the conclusion of the year's production of cones. There was no stunting of any of the leaves of the vegetative point, and the point was small in every case and clearly meristematic, as shown by the staining. Later in the season, where there were several staminate cones, they were nearly of the same size and apparently of about the same stage in development, so that they might easily be supposed to be of the same age and to result from a forking of the stem, as SACHS thought. The axes of the sympodium in such cases are shortened, so that the branches stand almost upon a level (*fig. 6*). Good figures are shown by WIELAND (21).

The ovulate strobili are generally formed singly, one a year from the vegetative point. The staminate strobili develop more rapidly and from a succession of vegetative points, but they reach maturity at about the same time. The two or three cases in which more than one

ovulate strobilus was formed agreed in the details of development with those of the staminate strobilus, and like them the several strobili may mature the same year. This is not infrequent in *Zamia floridana*.

The trunk of the cycads, and especially of *Zamia*, is capable of forming new tissue readily upon wounding, as COULTER and CHRYSLER (22) have shown. Since there is this power of forming secondary meristem, it may be that some of the cases of lateral shoots, produced upon the lower part of the trunk, are the results of wounding.

Since other cycads are known to produce more than one strobilus upon the trunk in the same year, the manner of growth of the stem in these forms may be the same as in *Zamia*. The staminate cone mentioned by DEBARY (10) had the same position as those of *Zamia*, with the vegetative point at the base of the cone, so that this case may be explained as a sympodium, and there may have been no stunting of the point as DEBARY thought. In view of these facts it seems probable that in all the other genera the branching is sympodial, except in the case of the ovulate plants of *Cycas*.

The successive pushing aside of the terminal strobili in *Zamia* (fig. 5) suggests comparison with Bennettitales, as described by SCOTT (17) and WIELAND (19), in which the strobili arise terminally on short, lateral branches, wedged in between the bases of the leaves. In the modern cycads, as illustrated by *Zamia*, the strobili finally occupy positions in the axils of the leaves, and the vegetative point crowns the stem at the end of the year's production of strobili.

## II. Microsporophylls and microsporangia

### DEVELOPMENT OF SPOROPHYLLS

The youngest staminate strobilus of *Zamia floridana* was obtained from material received from Miami, June 1, 1905. The leaves were removed from the crown of the plant and the small cone exposed, less than 1<sup>m</sup>m in length (fig. 7). The leaves in a vegetative point arise as small protuberances from the extreme base of the meristematic apex and incline toward each other, covering the point of growth (fig. 8). The strobilus, however, rises higher before showing any protuberances, and when these appear they rise acropetally and project at right angles to the cone (fig. 9). In the strobilus shown in fig. 10 the sporophylls are not much farther advanced than in fig. 7, but the strobilus has lengthened and there are more primordia of sporophylls.

The first protuberance of the sporophyll is brought about by a periclinal division of a hypodermal cell in the axial row (fig. 11), and later by the growth and division of the hypodermal cells and of layers beneath them (fig. 12). The outer layer divides anticlinally, but at the tip of the strobilus the divisions are also periclinal. Strobili of June 15 show further cell divisions in the young sporophyll and the appearance of rudimentary bundles (fig. 13). By July 8 the strobili have lengthened to 6 or 8<sup>mm</sup>. The new sporophylls appear toward the apex (fig. 9), but there is as yet no differentiation of sori. The strobili of July 25, which are 10 to 12<sup>mm</sup> long, exclusive of the peduncle, show fifteen or more sporophylls in a vertical row (fig. 14).

#### MATURE SPOROPHYLLS AND SPORANGIA

The mature microsporophylls of *Zamia floridana* are short-stalked and broaden outwards from their insertion, the sterile tip being thick and hairy. The microsporangia are borne upon the abaxial surface, and are grouped upon either flank, with the median region of the sporophyll bare. The sporangia extend to the margin but not beyond, so that there is no appearance of a "peltate expansion" of the sterile tip.

The microsporangia of *Ceratozamia mexicana* are borne upon obovate, cuneate sporophylls, whose sterile tips are produced into two horns spreading laterally. The sporangia cover the whole lower part of the sporophyll. The plates given by WARMING (7) show young stages of the sporophyll, when the sporangia are in two groups, one on either flank; but in the text he says that in further growth they become more numerous and spread toward the median region, so that at last this is covered.

In *Encephalartos villosus* the sporophyll has a very short stalk or is sessile; the tip is sterile and produced into a blunt point. The sporangia in the central sporophylls of the strobili cover the surface, but at the apex the outline of the sporangium-bearing area is notched in the center, showing a tendency toward a two-grouped arrangement. Single strobili of *E. Caffer* and of *E. Altensteinii* have been examined, and in both species throughout the median region of the cone the sporangia cover the sporophylls; but the lower sporophylls on the cone and those at the tip show fewer sporangia, and these are grouped into two lateral areas. The transition between these two conditions

can be traced in a series of sporophylls, which show the outline of the sporangium-bearing area becoming notched in the middle region above and below, and the notches gradually deepening until finally the middle region becomes bare. This series also occurs in *Dioon edule*; and in the one cone of *Macrozamia Miquelii* examined it was shown by the sporophylls at the base; but those of the central part and tip were entirely covered with sporangia.

The numbers of sporangia on a sporophyll range as follows: *Cycas circinalis* 700, *Encephalartos Caffer* 700, *Macrozamia Miquelii* 600, *Encephalartos villosus* 500, *Dioon edule* 200, *Zamia floridana* 24.

The sporangia on the outside of the sporophyll flank in *Zamia floridana* and *Ceratozamia mexicana*, also in Stangeria as recorded by LANG (16), are likely to have longer stalks than the sporangia on the central part, and are more rounded. In Ceratozamia they are so crowded that they become angular, often irregular in shape, and encroach upon one another (fig. 15); and, as in Stangeria, there are many hairs among the sporangia, arising from the sporophyll. In *Zamia floridana* and *Encephalartos villosus* there are no hairs in the region where the sporangia are borne.

The sporangia are definitely grouped into sori, arising from a cushion of meristematic tissue elevated slightly above the sporophyll (fig. 16). Two to five sporangia occur in the sori of Ceratozamia, Stangeria (according to LANG), *Macrozamia Miquelii*, *Encephalartos villosus*, and *E. Caffer*; two to four in *Cycas circinalis*; and seldom more than two in *Zamia floridana*. Into this soral cushion there is an extension of the vascular system, as in the synangia of Marattiaceae.

LANG (16) found the sporangia of Stangeria in every stage of development on the sporophyll, beginning at the center of each flank and extending toward the margin and middle region. He refers to WARMING's (7) figures of Ceratozamia to show that in that form the sporangia on a sporophyll are of the same age; but WARMING's text distinctly states that at first there are groups of twenty to thirty on each flank, and by further growth these spread so as to conceal the middle region. In my material of Ceratozamia, which was nearly all mature, the sporangia were about the same age; but a few on the edges and toward the center were a little younger.

In *Zamia* and *Ceratozamia* there is strong suggestion of the syn-

angial origin of the sorus. In the former, while the sporangia of a sorus are usually quite distinct, occasionally they are free for only about half their length (*fig. 17*); while in the latter this half-synangial character is more common (*figs. 18-20*).

One cone of *Zamia floridana* showed great variation in the number of sporangia on a sporophyll. The usual sporophyll (*fig. 21, a*) bore twelve on each flank, arranged in six pairs. Usually each flank had the same number of pairs, but this was not always the case. The number of sporangia ranged from two to forty-eight; and instead of being confined to the flanks, in several cases there were groups of sporangia on the middle region or even all the way across the sporophyll, approaching the condition in *Ceratozamia* (*figs. 21, b, c*). The number was likely to be less upon the sporophylls near the tip of the strobilus and upon those at the base. One sporophyll is shown in *fig. 21, d*, which suggests the megasporophyll in the position and number of sporangia; and it may be interesting to note that an ovulate cone furnished several sporophylls with two sporangia on each flank.

#### DEVELOPMENT OF SPORANGIA

Two papers by WARMING (6, 7) give the earliest information concerning the development of the microsporangium of cycads, the species investigated being *Ceratozamia mexicana* and *Cycas circinalis*. Later, TREUB (8) investigated *Zamia muricata* and LANG (16) *Stangeria paradoxa*, both giving an account of the sporangium from its early stages to its maturity, but neither obtained stages earlier than a several-celled sporogenous tissue.

A tangential section through the strobilus of *Zamia floridana* gives the best view of the origin of the sporangia (*fig. 14*); about three showing upon each lobe of the sporophyll in such a section, the youngest near the margin. The cells in the region where the sporangia arise stain more deeply than the rest of the tissue, and this meristematic condition extends below the sporangia for some distance. The epidermis here divides only anticlinally. In such a section as *fig. 14*, certain cells may be distinguished by their larger size, more deeply staining contents, and larger nuclei in which the chromatin is more prominent. This is the region which will give rise to the sporangia. In such meristematic groups a single hypodermal cell with large nucleus and deeply staining chromatin can be distinguished,

being larger than the surrounding cells and in an axial position (*fig. 22*); this cell is the archesporium. In sections parallel to the surface of the sporophyll this same cell can be distinguished from the surrounding cells (*fig. 25*), so that it is evident that it is the single archesporial cell which GOEBEL (11) predicted in reviewing TREUB's work. TREUB (8) failed to find in *Zamia muricata* less than four spore-gogenous cells, and LANG (16) concluded that the corresponding four cells in *Stangeria* represented the archesporium, which was "probably not a single cell." The archesporial cell divides usually by an anticlinal wall (*fig. 23*), and the two daughter cells lie side by side in a cross-section through the sporophyll; one cell is often larger than the other and divides earlier (*fig. 24*).

Occasionally the archesporial cell divides by a periclinal wall (*fig. 26*), suggesting the statement of BOWER (15) in reference to *Angiopteris*, in which the divisions are not always in the same direction in different sporangia. This exceptional division is shown also in a section parallel to the surface of the sporophyll (*fig. 27*).

The second division is anticlinal also, resulting in a hypodermal plate of four cells, only two of which are seen in cross-section.

The third division is periclinal, resulting in an outer and inner plate of four cells each (*fig. 24*), the outer plate being the primary wall cells, the inner plate the primary spore-gogenous cells. Further divisions of the spore-gogenous tissue are shown in *figs. 28–30*.

The wall is four to seven layers of cells in thickness, always thicker at the angles and in the region of the apex (*figs. 31, 32*); there is some small increase in the number of layers toward the center of the sorus also. The cells of the two layers adjacent to the tapetum are narrow and flattened (*fig. 33*), and later are crushed by the development of the spore-gogenous cells and the activity of the tapetum. The epidermal cells are thin-walled in *Ceratozamia* and *Zamia* up to the time of tetrad formation. At this time in *Zamia* they begin to show thickening along the crest of the sporangia, where the cells are deeper and narrower. Here the inner and vertical walls become thicker (*fig. 32*), so that by the time spores are formed these cells make a well-defined band along the crest of the sporangium; at the time the spores are ready to be shed this wall has become very thick. The new layers of the wall fill the cell until the lumen almost disappears. The

walls of the cells between the bands, *i.e.* those of the line of dehiscence, are at first very thin, but thicken up later, until they are almost as thick as those of the other cells.

The apex of the sporangium is composed of isodiametric cells (in cross-section) with uniformly thickened walls, beneath which is a group of cells which are thickened in the same way (*figs. 34-36*). From the apex, extending along either side of the line of dehiscence, the cells of the bands are longer, narrower, and have the inner and vertical walls thickened, the bands extending along the crest from the apex almost to the base of the sporangium.

The mature condition of the sporangium of *Encephalartos*, the only stage of this genus available, is much the same (*fig. 35*).

The older stages of the sporangium of *Ceratozamia* show the apical group of cells (*fig. 31*) projecting only slightly; and the band cells are little more thickened than the other cells of the sporangium, but are narrower. Cross-sections made from the younger stages (*fig. 37*) show in the wall a double line of cells running from the outside into the sporangial cavity, whose contents stain more deeply. These lie just beneath the line of dehiscence, and are therefore like a plate cutting into the wall from the surface. Just under the line of dehiscence in *Marattia* there are deeply colored mucilaginous cells which may aid in dehiscence; these slightly resemble those in *Ceratozamia*. Older stages of the wall do not show this plate; but there are occasionally several cells under the line of dehiscence, in which there is a great number of crystals. These are even more striking in *Zamia*, where early stages in their formation have not been seen (*fig. 38*).

At the time of spore-formation in *Zamia* there is also just opposite the cells of dehiscence, on the inner surface of the sporangium, a projection into the wall of the inner layers and of the tapetum (*fig. 38*). The cells between are flattened and out of shape, as if degeneration had begun. This has been seen in a number of sections, and may or may not be a normal occurrence. Enough sections have not been seen to make a definite statement as to the function of these cells, but it may be suggested that there is a breaking down of the walls at the time of dehiscence, which aids in the breaking open of the sporangium. This may be seen in *Danaea*, where of course there is no mechanical arrangement for dehiscence; and possibly

this breaking down of cells is combined in *Angiopteris* with the regular mechanical arrangement for dehiscence.

In *Zamia* the stomata are situated in about the same region of the sporangium as in *Stangeria*. They are especially numerous among the thinner cells of the lower part of the sporangium, and the guard cells in cross-section are only about one-half as high as the other cells. The walls of the guard cells and of the subsidiary cells are strongly thickened, and the whole apparatus resembles that found among xerophytic angiosperms. The stomata of *Ceratozamia* are not so deeply sunken; but in *Encephalartos* they are more deeply placed than in *Zamia* (*figs. 39, 41*).

In *Zamia* and *Ceratozamia* the tapetum (some of it at least) is derived from the sporogenous tissue. The blocking out of the cells and the irregular width of the layer, especially toward the base of the sporangium, suggests this, and in many cases can only be explained by the fact that there is a distinct transformation of sporogenous cells into tapetum. In many sections it is hard to distinguish the tapetum, but in the stage shown in *fig. 42* the nuclei are smaller and a little more elongated, and the chromatin stains more deeply. At the base of the sporangium the irregularity in the width of the tapetum is more striking. Usually there are one or two layers all around the sporogenous mass; but *fig. 43* shows four or five layers, dipping into the sporogenous tissue. The nuclei of the tapetal cells become divided and spindles (*fig. 42*) are occasionally seen, so that although there may be fragmentation, as LANG (16) states, there is also regular mitosis. The tapetum is a distinct layer up to the tetrad stage, when the walls disappear, and the mass of nutritive substance lines the sporangial cavity.

There is some sterilization of sporogenous tissue, for occasionally regions of tissue in the center of the sporangium or at the base, with extensions into the center, become sterilized, and, as has been mentioned, the cells near the base often appear to function as tapetal cells.

The division of the spore mother cell was observed in *Ceratozamia mexicana* (*fig. 44*). The spirem in the nucleus of the mother cell is large and clear, the chromosomes are thick and short, so that the chance for counting is unusually good. A count was made in 50 nuclei, resulting in 12 chromosomes in 46 cases, 11 in 3 cases, and 13

in one case. In most of these cases the 12 chromosomes were visible in one section 3 to 5  $\mu$  thick (fig. 45), but in a few cases the sections on either side had to be examined and drawings made to determine the actual number of chromosomes. This in every case changed what would have been a count of 6 or 8 to 12. GUIGNARD (12) found 8 chromosomes in the pollen mother cells of *Ceratozamia*; and OVERTON (13) found the same number in a count of the chromosomes in the endosperm cells of the same species. In the 25 nuclei of *Zamia floridana* counted, 12 chromosomes were found; and CHAMBERLAIN (27) gives the same number for *Dioon edule*.

Most of the stages in the development of the tetrad were seen in *Zamia*, and they followed closely the figures of JURANYI (4) and TREUB (8). In most of the tetrads the ring of cellulose was not seen all the way across the cell until the four daughter nuclei were clearly outlined and the cell plate between them had commenced to grow.

In *Zamia floridana* the output of spores per sporangium is about 500 or 600; in *Ceratozamia mexicana* 8,000; and in *Encephalartos villosus* 26,000. This makes the output per sporangium in the few species examined increase according to the number of sporangia to the sporophyll. The number of observations, however, is too small to furnish a safe basis for conclusions. Among the Marattiaceae the largest output is for *Kaulfussia* (7,850); and in *Isoetes* SMITH (18) estimates several hundred thousand spores in a single microsporangium.

#### THE MALE GAMETOPHYTE

The strobili of *Zamia* received from Florida the first of February were about ready to shed the pollen, which was in the three-celled stage. At the first observed division of the spore the prothallial cell is cut off, lying against the wall of the spore, with its inner face arching almost into the center of the spore (fig. 46). At the next division the generative cell is cut off (fig. 47), its nucleus being somewhat flattened and deeply staining. The tube nucleus is very large, stains less deeply, and lies generally in line with the other two.

A 10 per cent. sugar solution, sterilized and kept in sterilized dishes, was favorable for the growth of the pollen tubes. Some of the solutions containing pollen grains were kept in an oven at a temperature of 28° C., while others were kept at 21° C., the temperature of the room. In forty-two hours the tubes in the oven were two or

three times longer than those in the room (*figs. 48, 49*). These cultures gave an opportunity to study the breaking of the exine and the extrusion of the tube (*fig. 50*). In the unstained specimens the portion of the wall just touching the prothallial cell and passing down either side of it was more refractive than the rest. In stained material (*figs. 48, 49*) this had the appearance of a lighter wall just within the intine, and agrees with the "third wall" described by Miss FERGUSON (24) in *Pinus*. The starch in the growing tube is very abundant, massing about the tube nucleus and the generative cell (*fig. 49*); in longer tubes there is also a large quantity near the tip. The tube nucleus slowly migrates toward the tip, keeping just behind it. In *Zamia* there is no sign of the second prothallial cell mentioned by WEBBER (20), though many hundred microspores were examined.

Microspores of *Encephalartos*, which are very large, were especially favorable for a study of the divisions to the three-celled stage (*figs. 51, 52*), which are like those described for *Zamia*. Only one mitotic figure was seen, but it shows the size and shape of the chromosomes fairly well (*fig. 52*).

#### Summary

1. The stem of *Zamia floridana* is a sympodium, a vegetative point lying at the base of each strobilus. The staminate strobili develop one after the other from the successive vegetative points, each strobilus with a small circle of leaves, and all enclosed within the larger scale leaves of the first strobilus.
2. The youngest staminate strobili received June 1 showed the sporophylls arising in acropetal succession, by a periclinal division of a hypodermal cell, and later by the divisions of other hypodermal cells and layers beneath them.
3. The microsporangia cover the abaxial face of the sporophylls in *Ceratozamia mexicana*, and are grouped upon each flank in *Zamia*. In *Dioon*, *Encephalartos villosus*, *E. Caffer*, and *Macrozamia Miquelii* there are intergrades, where the sporangia cover the sporophylls of the central part of the strobilus, but are in two groups upon the sporophylls of the tip and base.
4. The microsporangia are grouped in sori (two to six sporangia in each sorus), which are raised on a cushion of tissue into which a vascular bundle passes.

5. In *Zamia* and *Ceratozamia* the sporangia of a sorus are sometimes free only half their length.

6. One strobilus of *Zamia floridana* had two to forty-eight sporangia on its different sporophylls, and some sporophylls in which the middle region was entirely covered.

7. Strobili of *Zamia floridana* received July 25 showed in tangential sections three or four developing sporangia upon either flank of the sporophyll, the youngest upon the margin.

8. The archesporium is a single hypodermal cell which usually divides first anticlinally, followed by pericinal divisions; the outer plate of four cells developing the wall layers, the inner plate the sporogenous tissue.

9. The wall of the mature microsporangium is composed of four to seven layers of cells, the cells of the two layers next the tapetum being tabular. The apex of the sporangium consists of thick-walled cells and beneath it there are isodiametric, thick-walled cells. Extending from the apex on either side of the line of dehiscence there is a band of thicker-walled cells, which suggests the structure of the sporangium of *Angiopteris*.

10. Beneath the line of dehiscence there is a plate of cells extending toward the center of the sporangium, which contain crystals; and in *Zamia* these cells seem to degenerate as the sporangium breaks.

11. Stomata occur on the microsporangium, the guard cells being deeply sunken and thick-walled.

12. The tapetum is derived from sporogenous tissue, at least in part, and the sporogenous cells at the base of the sporangium function as tapetal cells, which may project into the sporangial chamber. Sometimes, at least, the tapetal nuclei divide mitotically, for spindles were seen; and there are often two nuclei to a cell. The tapetum remains a distinct layer up to the tetrad stage.

13. There is some additional sterilization of sporogenous tissue in the sporangium.

14. The divisions of the spore mother cells conform to the descriptions of JURANYI and TREUB. The reduced number of chromosomes in *Ceratozamia* and *Zamia* is twelve.

15. The output of spores per sporangium for *Zamia floridana* is 500-600, for *Ceratozamia mexicana* 8,000, and for *Encephalartos*

*villosus* 26,000. Therefore the output per sporangium in these species increases according to the number of sporangia upon the sporophyll.

16. The pollen grains are three-celled at the time of shedding, the cells being the prothallial, the generative, and the tube.

This investigation was carried on under the direction of Professor JOHN M. COULTER and Dr. C. J. CHAMBERLAIN, of the University of Chicago.

SMITH COLLEGE  
Northampton, Mass.

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#### EXPLANATION OF PLATE X

(Unless otherwise stated, the figures are from *Zamia*.)

FIG. 1. Diagram of the crown; *a*, peduncle of ovulate strobilus of last year; *b*, young ovulate strobilus; *v*, position of the vegetative point.  $\times 1$ .

FIG. 2. Longitudinal section of two ovulate strobili; between them is the vegetative point (*v*) and the leaves (*l*) surrounding it; the upper portion of the strobilus is cut away.  $\times 13$ .

FIG. 3. Longitudinal section of ovulate strobilus; *v*, vegetative point; *l*, rudimentary leaves; two vascular bundles enter the strobilus.  $\times 23$ .

FIG. 4. Longitudinal section of staminate cone (June 14); *v*, vegetative point; *l*, rudimentary leaves.  $\times 23$ .

FIG. 5. Longitudinal section of three staminate strobili (July 15); *v*, vegetative point; youngest strobilus (*st*) is cut at an angle.  $\times 11$ .

FIG. 6. Diagrams: *a*, sympodium; *b*, sympodium with shortened internodes; *c*, sympodium with internodes shortened until strobili are on a level, as in *Zamia*.

FIG. 7. Longitudinal section of young staminate strobilus (June 1), showing primordia of sporophylls (*sp*) and leaves surrounding strobilus.  $\times 27$ .

FIG. 8. Longitudinal section of typical vegetative point and rudimentary leaves.  $\times 32$ .

FIG. 9. Longitudinal section of staminate strobilus (July 8), showing sporophylls.  $\times 23$ .

FIG. 10. Longitudinal section of staminate strobilus a little further advanced than in fig. 7.  $\times 32$ .

FIG. 11. Section of staminate strobilus of fig. 7, showing the periclinal division of a hypodermal cell which is the first stage in the development of a sporophyll.  $\times 650$ .

FIG. 12. Section of staminate strobilus, showing further divisions of hypodermal cells and layers beneath them to form sporophyll.  $\times 650$ .

FIG. 13. Longitudinal section of strobilus, showing elevation of sporophylls and rudimentary bundles passing into them.  $\times 80$ .

FIG. 14. Tangential section of staminate cone of July 25, showing sporophylls and position of young sporangia.  $\times 27$ .

FIG. 15. Cross-section of sporophyll of *Ceratozamia*, showing sporangia.  $\times 37$ .

FIG. 16. Cross-section of sporophyll of *Ceratozamia*, showing grouping of sporangia into sori and extension of bundles into them.  $\times 27$ .

FIG. 17. Cross-section of sporophyll of *Zamia*.  $\times 39$ .

FIGS. 18, 19. Three sporangia of *Ceratozamia* "united" at base.  $\times 95$ .

FIG. 20. Detail of fig. 19, showing union of sporangia at base.  $\times 500$ .

FIG. 21. Sporophylls from one cone of *Zamia floridana*: *a*, usual arrangement of sporangia; *b*, sporangia extending to middle region; *c*, sporangia extending across middle region; *d*, reduced number of sporangia.  $\times 2$ .

FIG. 22. Sporophyll of fig. 14, showing hypodermal archesporial cell.  $\times 920$ .

FIG. 23. Division of archesporium by an anticlinal wall.  $\times 920$ .

FIG. 24. The periclinal divisions resulting in inner and outer plates of cells.  $\times 920$ .

FIG. 25. Archesporial cell in a section parallel to the surface of the sporophyll.  $\times 920$ .

FIG. 26. Periclinal division of archesporial cell (exceptional).  $\times 920$ .

FIG. 27. First (anticlinal) division of archesporial cell in a section parallel to the surface of the sporophyll.  $\times 920$ .

FIG. 28. Further periclinal divisions succeeding the division shown in fig. 26.  $\times 920$ .

FIG. 29. Usual divisions following the stage shown in fig. 24; lower four cells of section are sporogenous.  $\times 920$ .

FIG. 30. Longitudinal section of sporangium in more advanced stage.  $\times 920$ .

FIG. 31. Longitudinal section of sporangium of *Ceratozamia*; tapetal cells marked by +.  $\times 180$ .

FIG. 32. Longitudinal section of sporangium of *Zamia*, showing thickened cells at apex and band of dehiscence.  $\times 95$ .

FIG. 33. Detailed drawing of part of sporangium: *w*, outer wall layers; *y*, two layers of tabular cells, which are crushed by the activity of the tapetum; *t*, tapetum; *s*, sporogenous cells.  $\times 800$ .

FIG. 34. Apex of sporangium of *Zamia*, showing thick-walled cells; isodiametric cells beneath them.  $\times 580$ .

FIG. 35. Diagram of sporangium of *Encephalartos*.

FIG. 36. Tangential section through sporangium of *Zamia* near apex, showing thick-walled cells and line of dehiscence (*d*).  $\times 180$ .

FIG. 37. Cross-section of sporangium of *Ceratozamia*, with plate of cells extending from beneath line of dehiscence to the sporangial cavity.  $\times 500$ .

FIG. 38. Cross-section of mature sporangium wall; *c*, cell containing crystals, just beneath the line of dehiscence; cells of inner layers of sporangial wall projecting into outer layers; *t*, tapetum broken down; *m*, microspore.  $\times 800$ .

FIG. 39. Longitudinal section of two sporangia of *Encephalartos*.  $\times 70$ .

FIG. 40. Surface section of stoma in the sporangial wall of *Encephalartos*; *g*, guard cells; *su*, subsidiary cells.  $\times 650$ .

FIG. 41. Cross-section of stoma of *Encephalartos*.  $\times 650$ .

FIG. 42. Detail of sporangium of *Ceratozamia*: *t*, tapetum; *sp*, sporogenous cells.  $\times 120$ .

FIG. 43. Longitudinal section of sporangium of *Ceratozamia* at the base; tapetal cells becoming more numerous by sterilization of sporogenous tissue.  $\times 180$ .

FIG. 44. First division of pollen mother cell.  $\times 1500$ .

FIG. 45. Nuclear plate in spore mother cell, showing 12 chromosomes.  $\times 1500$ .

FIG. 46. Male gametophyte of *Encephalartos* in 2-celled stage; *p*, prothallial cell; *a*, antheridial initial.  $\times 1500$ .

FIG. 47. Male gametophyte of *Encephalartos* in 3-celled stage; *p*, prothallial cell; *g*, generative cell; *t*, tube cell.  $\times 1500$ .

FIG. 48. Male gametophyte of *Zamia*, showing beginning of tube; lettering as before; *z*, "third wall," showing within intine.  $\times 1500$ .

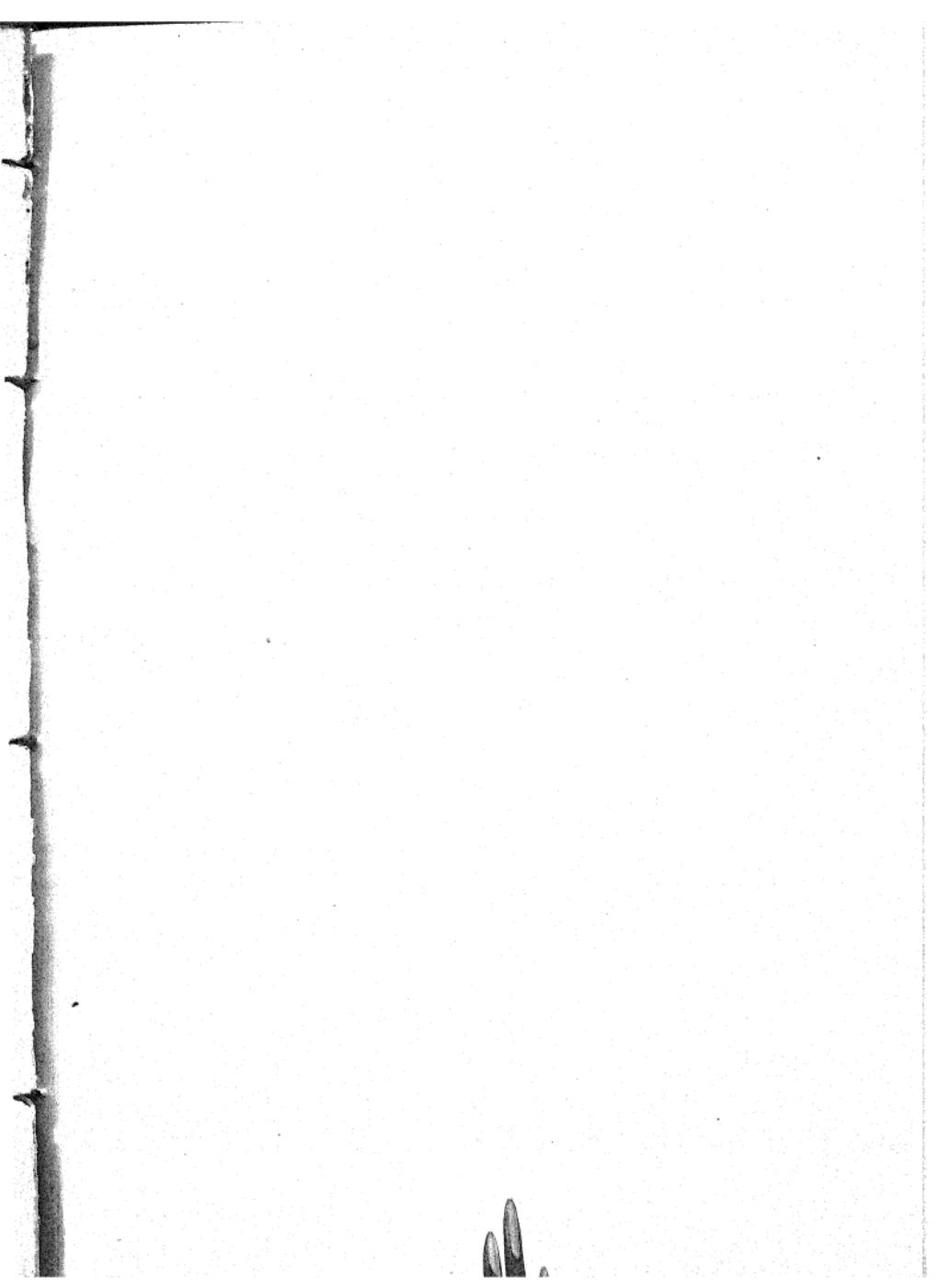
FIG. 49. Further development of pollen tube; tube nucleus passing into the tube; much starch present.  $\times 1500$ .

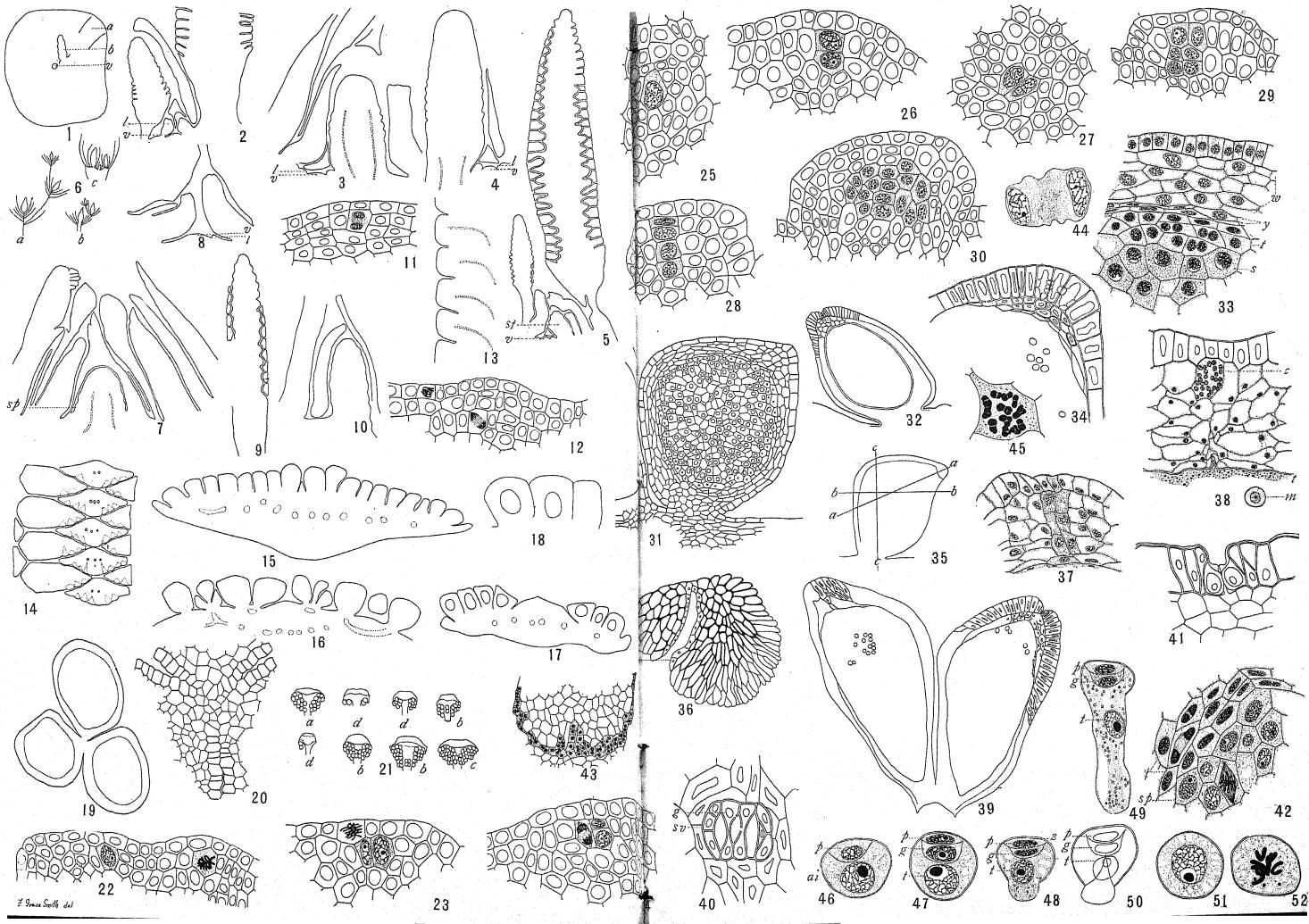
FIG. 50. Culture from *Zamia*, showing extrusion of tube and breaking of exine.  $\times 1500$ .

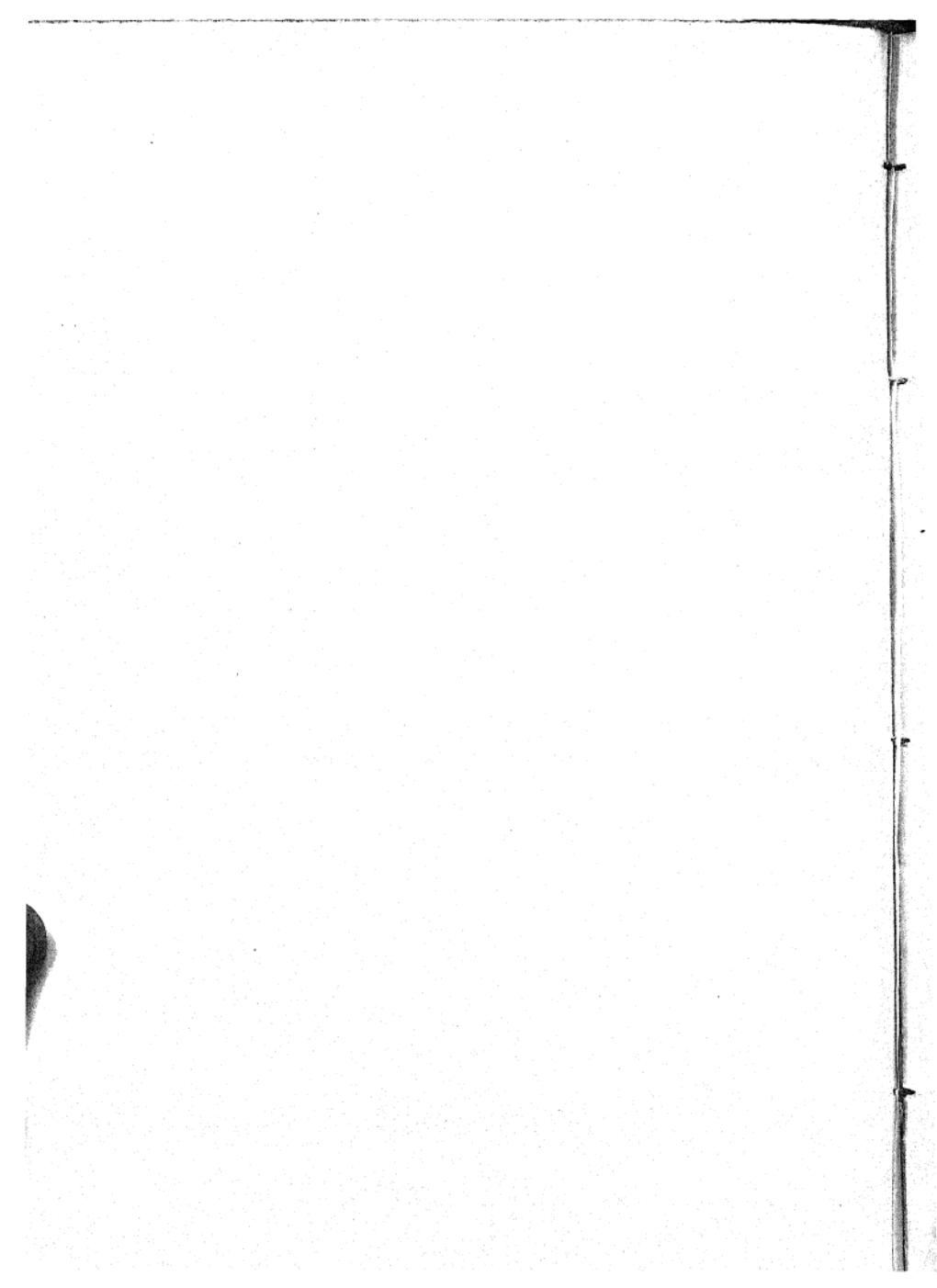
FIG. 51. Microspore of *Encephalartos* in resting stage.  $\times 1500$ .

FIG. 52. Microspore of *Encephalartos*, showing some of the chromosomes.  $\times 1500$ .

The magnifications given in the foregoing explanations are those of the original drawings, which have been reduced three-fifths in reproduction.







## BRIEFER ARTICLES

### CHARACTERS OF *PINUS*: THE LATERAL CONE

(WITH TWO FIGURES)

In *Trees and Shrubs* (1:209. 1905), under *Pinus Altamirani*, attention is called to the variation in the position of the young cone as follows: "Like some other Mexican pines with normally subterminal cones, it sometimes produces double nodes and lateral cones and pseudo-lateral young cones occasioned by a short incomplete midsummer growth." The three possible positions of the young cone are here found in the same species, and suggest the investigation of characters which have been considered by some authors to be of weighty significance in the determination not only of species but of comprehensive sections of the genus.

In comparing the winter bud of *Pinus Strobus* with that of *P. rigida* or *P. Banksiana* (*divaricata*), two distinct forms are seen, the former predicting a single internode for the shoot of the following spring, the latter two or more. A *uninodal* shoot, when developed, consists of a shorter leafless base (which bears the staminate flowers when they are present) and a longer portion bearing the foliage and terminating in a node of buds, a *terminal* bud, and a verticillate group of *subterminal* buds about its base. A *multinodal* shoot, in addition to this, comprises one or more inner internodes, each clearly defined by a leafless base at one end and a node of lateral buds at the other (fig. 1). The pistillate flower, the future cone, takes the place of one of the subterminal or lateral buds. On uninodal shoots its position is necessarily subterminal; on multinodal shoots it may be either subterminal or lateral or both, even on the same shoot.



FIG. 1

Evidently the characters implied in the "subterminal and lateral cones" of authors would have a wider application and a more accurate significance if expressed in terms of the uninodal and multinodal vernal shoots, since these last characters are present on young or sterile as well as on fertile trees, while the lateral and subterminal conelets often coexist on the same branch. "Subterminal cone" is an unfortunate term. No distinction has been made by authors between the immature cone of the first season, which may be either subterminal or lateral, and the ripe cone of the following season when it is invariably and inevitably lateral; therefore "conelet," proposed by MOHR exclusively for the cone of the first year, will be used here.

After the vernal growth is clearly defined and the flowers have been pollinated, it often happens that a summer growth takes place that was not apparent in the winter bud. This growth, in the summer, differs from the spring growth not only in its less development but also in its *green* bracts, which, not being required for the protection of the winter bud, assume more or less completely the size, color, and character of the primary leaf; and at the end of the season, when the bracts have withered or fallen away, this summer shoot can be recognized by its shorter leaves. This growth, of course, does not affect the status of those conelets whose lateral position has been established already on the spring growth, but it reduces subterminal conelets to a quasi-lateral position and converts a uninodal into an imperfect multinodal shoot.



FIG. 2

This summer growth is quite common and may occur on any pine. On most species it is merely sporadic, appearing here and there on vigorous branches, more commonly on younger than on older trees; on a few species it is usual and characteristic, at least in youth. Of these latter *P. Bungeana*, the Chinese nut pine, is a very perfect example, and its hardiness in this latitude offers an excellent opportunity for the study of this peculiarity (fig. 2). The three positions of the conelet, corresponding to the uninodal, multinodal, and summer shoots, may be conveniently distinguished respectively as subterminal, lateral, and pseudo-lateral conelets.

They have been described, to avoid ambiguity, without qualification and without considering the variations and exceptions that arise from the complex influences, internal and external, temporary or permanent, that may modify the development of the annual growth. As a matter of fact, while the characters under consideration may be potential in a species, they are not always invariable or consistent.

A multinodal shoot, at the lateral nodes, may put out conelets or branchlets or both; either may be absent or be represented by a bud which may or may not develop the following year; or there may be nothing whatever to indicate the lateral node except the leafless base of the internode beyond. Again, the winter bud is an incipient branchlet, the beginnings of a growth to be continued in the spring; the degree to which the bud is developed, at the end of the summer, is subject to the vicissitudes of all growth. The bud destined to produce a multinodal shoot may be so far advanced as to show its purpose at a glance, or its future development may be latent and concealed to a greater or less degree. Multinodal pines often produce uninodal shoots, and this apparent inconsistency becomes more frequent with increasing age, so that individuals of *P. rigida*, and allied species, may be found which are nearly or absolutely uninodal. Uninodal pines, on the contrary, are more constant. The summer shoot, however, may occur on any species, and may show in autumn various degrees of development which may amount to no more than a slight elongation of the bud or may form a conspicuous tuft of leaves on the end of the branchlet. From its very nature the summer shoot must be regarded as evidence of local or temporary vigor rather than as a specific character, and this is true even of those pines where the development of the summer shoot has almost diagnostic value. *P. Bungeana* and *P. Gerardiana* are credited by authors with "lateral cones" on account of the persistency of this summer growth; but specimens in the herbaria at Kew, Paris, and the Arnold Arboretum show conclusively that their pseudo-lateral conelets partly or completely disappear in mature trees.

Taking the genus as a whole, there seems to be every gradation between the two extremes, the conelet exclusively lateral and the conelet exclusively subterminal. These considerations point to the conclusion that the difference implied in the "subterminal and lateral cones" of authors is one of degree rather than of kind, and however valuable the lateral conelet may be, when it is present, for the determination of species, it is not available, on account of its inconstancy, for broader classifications. It is therefore evident that a herbarium specimen, so far as it shows these characters, may not represent the normal behavior of a species, and such a specimen

may be misleading. In *Jour. Linn. Soc.* (35:601) "a specimen of *P. Montezumae* containing leaves 12-14 inches long and others 5 inches long on the same branch" is cited as if it were unusual. It is evidently the arrested growth of a summer shoot that bears the shorter leaves.

In *Bull. Torr. Bot. Club* (30:108), *Pinus cubensis anomala* Rowlee, a new variety, is described with "bracts 8<sup>mm</sup> long, green on the young shoot," and further on, "the reversion of the scales to what is generally considered the primitive form of the primary leaf of the pine is an exceedingly interesting phenomenon." This again, as the specimen shows, is the summer shoot in its usual form, and its counterpart has been observed in a great number of species.

*P. teocote* Sch. and Deppe, is described as a pine with a lateral cone. The summer growth, which is quite frequent on this species, as well as on other Mexican pines, is responsible for this error, as the species bears normally subterminal conelets.

*P. contorta* is placed in ENGELMANN'S *Revision of the genus Pinus* (*Trans. St. Louis Acad.* 4:177) in his section PONDEROSAE, though not without hesitation (l. c. 182), on account of its subterminal cone. It is nevertheless a true multinodal pine, bearing cones on both lateral and terminal nodes.

In his recent work *Wald- und Parkbäume* (1906) HEINRICH MAYR makes a distinction between true and false nodes (*echte Quirle* and *Scheinquirle*), which may be recognized respectively by the presence or absence of bud scales; "subterminal and lateral cones" are explained in terms of these nodes. His sections PINASTER and JEFFREYA produce cones at the true nodes, his section MURRAYA at the false nodes. Apparently these characters are held to be invariable, and no allowance is made for the appearance of cones at both forms of the nodes on the same species. Only on this supposition can the anomalies that appear in MAYR's classification of the pitch pines be explained.

*Pinus rigida* is in his section MURRAYA; while *P. serotina*, which does not differ from *P. rigida* in the characters under consideration, is in his section JEFFREYA. *P. halepensis* is in PINASTER, while *P. Brutia* is in MURRAYA. *P. Sabiniana*, *P. Coulteri*, and *P. caribaea* which, so far as they are affected by these characters, belong in MURRAYA, are all in JEFFREYA. These inconsistencies may all be laid to specimens which happen to bear subterminal conelets instead of the characteristic lateral ones. *P. chihuahuana*, which requires three years in which to perfect its cone, is found in the section MURRAYA. Here his distinction between true and false nodes, if it is reliable, should have prevented MAYR from mistaking the conelet

of the second year for a lateral conelet. The species is uninodal and bears subterminal conelets.

It is not the purpose of this article to discuss the merits of MAYR'S very interesting arrangement of *Pinus* except so far as it involves the false node and its cone, on which his section *MURRAYA* is mainly founded. It serves to illustrate, however, the elusive character of the "lateral cone," which should be regarded as an incident, more or less persistent, in the life of a pine, rather than as an invariable character.—GEORGE RUSSELL SHAW, *Arnold Arboretum, Jamaica Plain, Mass.*

### THE GENUS ALISMA IN NORTH DAKOTA

(WITH ONE FIGURE)

As the basis for this investigation I have used BUCHENAU'S monograph.<sup>1</sup> According to his analytical key, *Echinodorus* has 6, 9, 12, or more stamens, and the carpels are arranged in a dense head; while *Alisma* is characterized by 6 stamens, and has the carpels placed in a circle. Consequently he has changed the name *Alisma tenellum* Martius to *Echinodorus tenellus* (Mart.) Buchenau. He refers all the other forms of *Alisma* to *A. Plantago* L. and has divided this species into three varieties: var.  $\alpha$  *Michaletii* Aschers. et Graebn.; var.  $\beta$  *arcuatum* (Michalet) Buchenau; and var.  $\gamma$  *parviflorum* (Pursh) Torr. He says that var. *arcuatum* "is distinguished by many special small characters" from the other varieties. As will be found below, some of these differences are most conspicuous and give to the plant a peculiar aspect, and the distinguishing characters are constant and extend to all parts of the plant. I have therefore restored this variety to its former specific rank, and believe that the following will be an acceptable synopsis:

1. Pedicels forming with the scape an angle of 45° or less; styles erect, longer than the ovules . . . . . *A. Plantago (aquatica)* L.
2. Pedicels forming with the scape an angle of 90° or more; styles bent outwards in a hook, shorter than the ovules . . . . . *A. arcuatum* Michalet.
  1. *ALISMA PLANTAGO* L., *vide* Engler's *Pflanzenreich*, l. c.—Scapes generally solitary (seldom two), 15–100<sup>cm</sup> high, erect; strong, slender, fibrous, as is the whole plant, except when very young. Leaves bright green, ovate to lanceolate, with an acute apex and a rounded or cordate sometimes tapering base; petioles 2–35<sup>cm</sup> long; the blades usually 7-ribbed, 5–17<sup>cm</sup> long, 3–9<sup>cm</sup> wide. Inflorescence a strict, large, loose, pyramidal panicle of majestic appearance, its lower part raised above the level of the leaf

<sup>1</sup> BUCHENAU, FR., *Alismataceae*. Engler's *Pflanzenreich* 4: no. 15. pp. 66. 1903.

tops, with a pointed, fastigiate apex. Pedicels verticillate in five or six rows, the erect rays forming secondary and tertiary verticils, each verticil subtended by three lanceolate, acuminate bracts. Petals 2-4<sup>mm</sup> long or smaller, white with yellow unguis, obovate, often with two small indentations. Sepals striate with five pairs of "nerves," oval, with white hyaline margins. Stamens six, twice as long as the ovules. Styles numerous, erect, longer than the ovules. Nuts not joined inwardly, thus leaving the center of the receptacle uncovered.—Growing in wet or dry mud.

Var. *PARVIFLORUM* (Pursh) Torr. Fl. N. United States 1:382. 1824.—*A. parviflora* Pursh, Fl. Am. Sept. 1:253. 1816. *A. trivialis* Pursh, l. c.—Flowers small, 3-4<sup>mm</sup> in diameter. Peduncles slender, thin, often bent. A broad-leaved form of this is *A. Planiago americanum* in Roem. et Schult. Syst. 7:1598. 1830 = f. *LATIFOLIUM*. A representative of this variety with lanceolate leaves (= f. *STENOPHYLLUM*) is *A. Geyeri* Torr. in Rep. Upper Miss. 162. 1843, according to BUCHENAU, though the description in RYDBERG'S *Flora of Montana* (p. 19) would rather identify it with *A. arcuatum* Michalet.

Var. *MICHALETII* Aschers. et Graebn. Synops. Mitteleur. Flora 1:382. 1898.—*A. major* S. F. Gray, Nat. Arr. Brit. Pl. 2:216. 1821.—Flowers larger than in the preceding variety. Petals twice as long as the sepals. Peduncles stouter as a rule, strict.

Forma *LATIFOLIUM* Aschers. et Graebn. l. c. 383.—*A. latifolium* Gilib. Fl. Lith. 5:222. 1781. *A. Plantago latifolium* Kunth, Fl. Berol. 2:295. 1838.—Leaf-blades usually large, ovate, acute, with rounded or cordate (more seldom attenuate) base.

Forma *STENOPHYLLUM* Aschers. et Graebn. l. c. 383.—*A. lanceolatum* With. Bot. Arr. Brit. Pl. ed. 3. 2:362. 1796. *A. ranunculoides* All. Fl. Pedem. 1:243. 1785. *A. Plantago lanceolatum* Kunth, Fl. Berol. 2:295. 1838. *A. major*  $\beta$  *lanceolata* S. F. Gray, l. c.—A smaller plant. Leaf-blades broadly or narrowly lanceolate, acute (sometimes with a repand-attenuate base). This is a form produced by insufficient nutrition of the plant.

*A. Plantago* L. grows over the whole temperate part of North America, where all the different enumerated varieties and forms will be found; var. *Michaletii* extends its range also over Europe, Asia, and northern Africa.

2. *ALISMA ARCUATUM* Michalet, Bull. Soc. Bot. France 1:312. 1854.—*A. Plantago*  $\beta$  *arcuatum* (Michalet) Buchenau, Ind. Crit. Abh. Nat. Ver. Bremen 2:34. 1871. *A. Plantago*  $\beta$  *decumbens* Boiss. Fl. Orient. 5:9. 1884.—Scapes two to four (usually three), growing out one by one, and thus showing the highest number late in the season, always unequal in

length according to age, commonly bent in different shapes, often like an S, adscendent, strict or procumbent, 6–60<sup>cm</sup> high; thick, fleshy, fragile, like the whole plant, when young, tough and leathery when old. Leaf-blades varying from broadly lanceolate to linear, tapering at both ends, usually 5-ribbed, 2–15<sup>cm</sup> long, 0.5–2<sup>cm</sup> wide, somewhat glaucous; petioles 3–35<sup>cm</sup> long, the length differing very much between the petioles of the same plant. The lowest part of the petiole is dilated, but only half as much as in *A. Plantago*, and with a narrow, scarious margin (*A. Plantago* has the lowest part of the petiole very much dilated and broadly scarious-margined). The inflorescence has generally a more rounded outline and apex than that of *A. Plantago*, and some or all of the leaves reach higher than the top. (At the late fruiting season the inflorescence of one of the scapes will generally reach above the top of the longest leaf.) Pedicels verticillate in two to six rows, the shorter generally thicker than the others, horizontal (or sometimes drooping), forming secondary verticils, but seldom tertiary, each verticil subtended by three lanceolate, acuminate bracts. Petals 1–2<sup>mm</sup> long, light rose-colored, with the yellow spot of the unguis comparatively much larger than in *A. Plantago*, 1.5 times longer than the sepals, the upper margin often more or less fimbriated. Sepals striate, with seven pairs of "nerves," oval, with deeply rose-colored hyaline margins, often giving the predominant color to the flower. Stamens six, of the same length as the ovules. Styles numerous, bent outwards in a hook, shorter than the ovules. Nuts joined inwardly, and thus covering the center of the receptacle.—The species is subdivided as follows:

Var. *pumilum* Prahls, Kritische Flora 2:204. 1890.—*A. Plantago arcuatum minimum* Buchenau, l. c. *A. Plantago pumilum* Notte in Sonder, Flora Hamb. 210. 1851.—With usually a single verticil (more seldom two).

Var. *lanceolatum* (Buchenau) Lunell.—*A. lanceolatum* et *A. Plantago lanceolatum* auct. div.—Leaf-blade elliptic to lanceolate (seldom linear-lanceolate), acute, tapering at both ends. This is the usual terrestrial or emersed form, growing under exactly the same conditions as *A. Plantago*, and mixed with it. (Fig. 1.)

Var. *angustissimum* (Aschers. et Graebn.) Lunell.—*A. Plantago arcuatum angustissimum* Aschers. et Graebn. l. c. p. 384. *A. natans* Poll. Hist. Pl. Palat. 3:319. 1777. *A. Plantago angustissimum* DC. Fl. Franc. ed. 3. 5:312. 1815. *A. Plantago graminifolium* Wahl. Fl. Ups. 122. 1820. *A. graminifolium* Ehrh. in Steud. Nom. 1:26. 1821. *A. angustifolium* in Opiz Böhme's Phan. u. Crypt. Gew. 48. 1823. *A. graminea* Gmel. Fl. Bad. 4:256. 1826. *A. Loeselii* Gorski, Eichw. Nat. Skizze Lith. 127. 1830. *A. longijolium* Presl. Sommer Königr. Böhmen 15:xlvi. 1847.

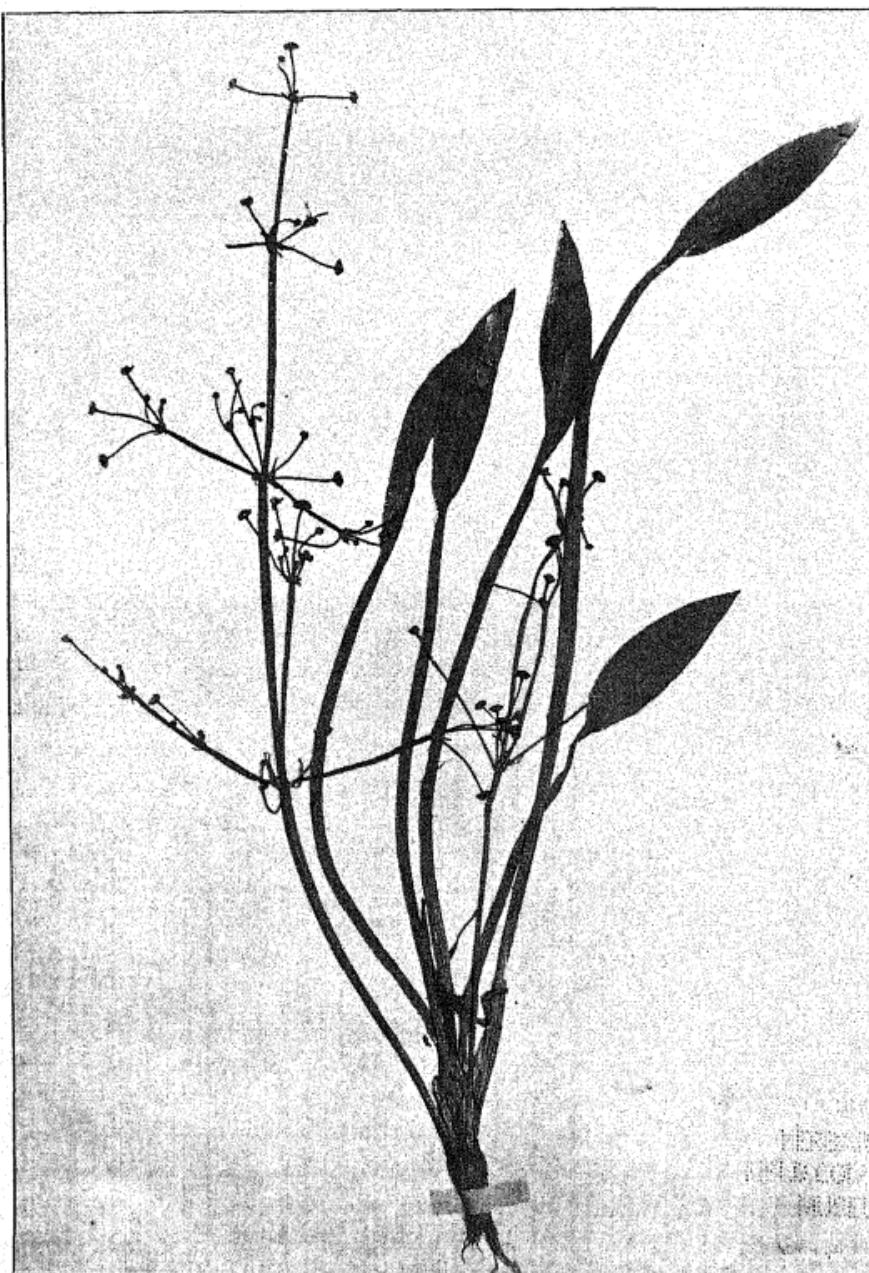


FIG. 1.—*Alisma arcuatum lanceolatum*.

*A. arcuata graminifolia* Casp. Schrift. Phys. Okon. Gesellsch. Königsberg 25:ii. 1884. *A. arcuata aquatica* Celak. l. c. 417.—Totally or partly submersed. All or most of the leaves swimming, linear, from a few to 100<sup>cm</sup> long, without blades (or one, or a few of the leaves with a linear-lanceolate blade, either swimming, or on a stiff petiole reaching above the surface of the water). Inflorescence more or less emersed or even submersed. Where the water is getting lower, it passes into var. *lanceolatum*, and plants with linear leaves without blades (phyllodes), within three or four weeks, with the gradual lowering of the water-level or disappearance of the water, have been transformed into typical specimens of var. *lanceolatum*, according to repeated observations by myself and others.

According to BUCHENAU, *A. arcuatum* is distributed only over central Europe, northern Africa, and western Asia (to Persia and western Siberia). To this belt of territory can now be added North Dakota, where I have found var. *pumilum* along Oak Creek, near Bottineau, and the other two varieties at Leeds and York. They have been collected during the months of July and August.

In the above presentation some additions of characters have been made to the original descriptions, and also some modifications thereof, to conform with conditions existing in North Dakota; but none of them has changed the essential features of the species.—J. LUNELL, *Leeds, North Dakota.*

## CURRENT LITERATURE

### BOOK REVIEWS

#### *Handbook of Dendrology*

The first volume of SCHNEIDER's work is now complete and it must be judged a very important addition to dendrological literature.\* It contains the families from Salicaceae to Pomaceae, and therefore deals with some of the most difficult families and genera of ligneous plants. The treatment is very exhaustive and an immense amount of information has been crowded into a comparatively small space. This could not be done without some disadvantage, however, for the numerous abbreviations make the perusal of the book somewhat difficult until one has become familiar with the shortened technical terms and the whole arrangement of the book. At the first glance it becomes apparent that the work is not a mere compilation, but based on original and thorough studies. The author shows himself well versed in botanical literature, both the older and the more recent, and must have examined a vast amount of herbarium material, for he quotes freely herbarium specimens, particularly in the more critical and in the new and more recently published species; in regard to *Crataegus*, for instance, he remarks that he had examined about 2000 specimens of American species.

The general treatment resembles somewhat that of ASCHERON and GRAEBNER's *Mitteleuropäischer Flora*, and, as in that work, much attention has been paid to varieties and hybrids, which of course is to be expected in a book devoted chiefly to cultivated plants. The arrangement of the species under dichotomous keys facilitates determination; some large genera are preceded by special keys, *e. g.*, there is a key to determine the 65 species of oaks from the foliage, and one to determine the 62 species of willows from flowering specimens without leaves. A great help for the determination are the numerous clear and well-reproduced illustrations, mostly representing analyses; according to the numbering they are 460, but each consists of 10 to 20 separate drawings, chiefly by the author himself. Not only are the species actually in cultivation described, but also related species not in cultivation and species likely to be sooner or later introduced are mentioned and more or less fully described, so that in several cases the treatment amounts really to a short monograph on the genus in question. Many new species and varieties, chiefly from China, but also from North America, appear here for the first time, and of course a large number of new combinations have incidentally been made. The nomenclature is that of the Vienna Congress; unfortunately

\* SCHNEIDER, CAMILLO KARL, *Illustriertes Handbuch der Laubholzkunde; Charakteristik der in Mitteleuropa einheimischen und im Freien angepflanzten angiospermen Gehölz-Arten und Formen mit Ausschluss der Bambuseen und Kakteen.* Band I. 8vo. pp. 810. figs. 460. Jena. 1904-1906.

the first parts of the book were published before that congress, and in these, therefore, a somewhat different nomenclature had been used, but the deviations from the Vienna rules are corrected at the end of the volume.

There is much in the book of great importance to American botanists. Genera like *Philadelphus*, *Cercocarpus*, *Holodiscus*, *Spiraea*, *Prunus*, *Amelanchier*, *Crataegus*, and other American genera of *Saxifragaceae*, *Drupaceae*, *Rosaceae*, and *Pomaceae* are exhaustively treated and many valuable notes on the affinities, the systematic value, and the conception of American species are included. It would of course be impossible to give here further details of the author's treatment of American species and of his notes and remarks, nor would it be of much use to select at random a few of them.

The book will prove not only indispensable to every student of cultivated trees and shrubs, but will also be of great importance to every botanist engaged in the study of the American, European, and Asiatic floras. In fact the work will amount almost to a representation of the whole ligneous flora of the temperate and colder regions of the northern hemisphere when it is completed, and it is to be hoped that the following volume or volumes will show the same thorough and careful treatment as the one now before us.—ALFRED REHDER.

#### Errera Botanical Institute

Two sumptuous volumes have recently appeared, containing the papers originating in this Institute.<sup>2</sup> As a publication the *Recueil* was planned by the late Professor LÉO ERRERA in 1902, and the first four volumes were intended to contain all the work done before that date in the botanical institute of which he was the inspiring director. But of these four only volume 1 is now first published. Volume 5 appeared in 1902 and contained papers prepared in 1900–1901. Volume 6 continues the series, under the editorship of Professor MASSART, the successor of the lamented founder, whose name the institute now bears.

In volume 1 as a frontispiece will be found an excellent photogravure of Professor ERRERA; ground plans of the institute are inserted; there are eleven papers by ERRERA himself, mostly reprinted, bearing on glycogen in the plant kingdom, beginning with his thesis in 1882 and closing with the posthumous and incomplete papers which have already been noticed in this journal.<sup>3</sup> Other reprints are:

The titles of three papers and the text of one, by ÉMILE LAURENT, all dealing with the physiology of yeasts, and another on the formation of starch from organic solutions; GEORGES CLAUTRIAU'S paper (1895) on glycogen in fungi,

<sup>2</sup> Recueil de l'Institut botanique (Université de Bruxelles) publié par L. ERRERA. Tome I, avec cinq planches. Glycogène, amidon et autres réserves non azotées.

Recueil de l'Institut botanique Léo Errera (Université de Bruxelles) publié par JEAN MASSART. Tome VI, avec vingt-sept figures dans le texte et vingt-trois planches. Bruxelles: Henri Lamertin. 1906. Each Jr. 25.

<sup>3</sup> BOT. GAZETTE 41:370. 1906, and 43:79. 1907.

and another on the hydrocarbon reserves (1899); and N. ENSCH's researches on glycogen in Myxomycetes (1899).

Volume 6 likewise consists chiefly of reprints, opening with MASSART'S memoir on irritability (1902); MOLLE'S account (1902) of an alkaloid, cliviine in *Clivia miniata* Benth.; ERRERA'S discussion (1903) of the limit of minuteness of organisms, and (1905) of the struggle for precedence and inhibitory action;<sup>4</sup> JOSEPHINE WERY'S experiments (1904) on the attraction of bees by flowers;<sup>5</sup> VAN RIJSSELBERGHE'S physico-chemical paper (1905) on the properties of mixed solutions and the physiological determination of their osmotic pressure; JACQUEMIN'S microchemical study (1905) of the localization of alkaloids in Leguminosae; ERRERA'S incomplete work on hygroscopicity as the explanation of ELFVING'S "physiological action at a distance" (of which more below), and a preliminary note suggesting that the form and development of leaves are related to atmospheric electricity; finally MALTAUX and MASSART'S paper on the chemical stimuli which affect the process of cell division.

ERRERA'S paper on hygroscopicity has been edited by COMMELIN from manuscript written in 1891, at which time ERRERA'S experiments on the curvatures observed by ELFVING in Phycomyces led him to ascribe them to hydrotropism; but these results were not published in full. Lately he had felt that it was necessary to reinvestigate the matter in the light of the new discoveries in radiation, and on this task he was engaged in 1905 at the time of his death. His notes and conclusions, so far as they could properly be used, have been incorporated by the editor; but the paper remains a fragment—yet a valuable one.

The volumes show the tremendous activity inspired by this genial *savant*, whose loss is felt most keenly. It is to be hoped that the Institute will continue its fine record under its new energetic director.—C. R. B.

#### Water relations of desert plants

LIVINGSTON has issued as publication 50 of the Carnegie Institution an account of studies made chiefly in the summer of 1904,<sup>6</sup> at the Desert Botanical Laboratory. He examined the soil moisture, atmospheric conditions, and the evaporation from water surfaces and plants. These studies are a worthy contribution to the solution of the problem of water relations of plants in the desert, in which many have interested themselves; but the end is not yet.

The soil of Tumamoc Hill, on which the laboratory is situated, was found to have a high retentive power, and to contain, even at the end of the dry season, 12–15 per cent. of water (by volume) at a depth of less than 40<sup>cm</sup>, an amount quite adequate to the needs of the desert plants. The water penetrates deeply

<sup>4</sup> See BOT. GAZETTE 41:221. 1906.

<sup>5</sup> See BOT. GAZETTE 40:476. 1905.

<sup>6</sup> LIVINGSTON, B. E. The relation of desert plants to soil moisture and to evaporation. Imp. 8vo. pp. 78. figs. 16. Publication no. 50. Carnegie Institution, Washington, D. C. 1906. 40 cents.

and rapidly along rock faces, and its real evaporating surface soon lies below the actual surface, the upper dry layers acting like a dust mulch. The seedlings send roots downward very rapidly and are slow in developing foliage until the root system is extended deep. The sap of cacti is found to have an osmotic pressure not notably higher than that of many mesophytes; but the gums, etc., in their cells, which do not sensibly raise osmotic pressure, hold back water. An evaporimeter, consisting of an exposed tube of porous porcelain filled with water and connected with a burette, enables ready measurement or automatic record of the effect of atmospheric conditions on evaporation, and by calibration the measurements may be converted into evaporation rate per surface unit of free water. Experiments show that no clue to real facts is given a study of transpiration from plants in a closed chamber,<sup>7</sup> because of the striking effects of air currents. Relative transpiration, i. e., the ratio of evaporation from a plant to evaporation from the same surface of free water, is proposed as the best way of expressing the facts. The highest relative evaporation noted was 0.785 and the lowest 0.008. Observations indicate a "physiological regulation" of transpiration, in which air temperature is apparently the dominant factor.

Throughout, the work shows the purpose of the author to grapple with the problem as a physical one; only here and there a phrase survives, such as "the absorptive power exerted by the plant," that recalls the less modern attitude. Surely it is only by conceiving the plant, the air, and the soil as a system, within which exchanges occur under very definite and definable physical conditions, that we can hope to state these exchanges intelligibly. Why not modify the evaporimeter by putting the foot of its water column into the soil and then try to state the changes within that system? When that can be done, there is hope for plant evaporation, and not till then.—C. R. B.

#### MINOR NOTICES

**Key to woody plants.**—In 1904 WIEGAND and FOXWORTHY published a very useful *Key to the genera of woody plants in winter*, including the genera with hardy representatives found growing wild or in cultivation within the state of New York. The first edition was exhausted, and a second one has now appeared.<sup>8</sup> The text has been revised, but not much enlarged, two or three genera have been added, and the keys to the conifers have been considerably expanded.—J. M. C.

**Food for plants.**—A new edition of a booklet under this title by HARRIS and MYERS is edited and published by WM. S. MYERS, who is now devoting his time and energy to the nitrate of soda propaganda. It goes without saying that the term "food" as used applies to the mineral salts which yield nitrogen and phosphorus. The book is a queer mixture of general information for the curious,

<sup>7</sup> Cf. CANNON's polymer method, Bull. Torr. Bot. Club 32:575. 1905.

<sup>8</sup> WIEGAND, K. M., and FOXWORTHY, F. W., A key to the genera of woody plants in winter. Second edition. pp. 33. Ithaca, N. Y.: The authors. 25 cents.

results of experimental cultures, directions for raising various crops with appropriate fertilizers, business maxims, a plea for good roads, etc. It has neither coherence nor apparent object beyond advertising under the guise of a handbook.—C. R. B.

**Botanical literature.**—Section M of the International Catalogue of Scientific Literature, devoted to botany, was published in July by the Royal Society of London. It contains the literature for 1904, and some belated entries for the preceding three years.<sup>9</sup> The volumes are improving in comprehensiveness and accuracy. Certainly no research laboratory can do without them.—C. R. B.

**Volatile oils.**—The semi-annual report of SCHIMMEL & Co., dated October-November 1906, contains an unusually extensive statement of the trade conditions respecting the volatile oils and the plants which produce them. Fifty pages also are devoted to a summary of recent researches on terpenes and the terpene derivatives.<sup>10</sup>—C. R. B.

**Genera Siphonogamarum.**—The ninth fascicle of DALLA TORRE and HARMS<sup>11</sup> list of the genera of seed plants continues the general alphabetical index of names, the last entry being Diplopeltis.—J. M. C.

#### NOTES FOR STUDENTS

**Galvanotropism of roots.**—Two studies on this topic appeared almost simultaneously last autumn. SCHELLENBERG investigated the influence of salts on the direction of growth of the roots of peas,<sup>12</sup> using roots of seedlings grown to a length of 3-4 cm in moist sawdust and then fixed vertical in very dilute solutions of various salts, with cotyledons exposed. The experimental vessel with the solution was connected by filter-paper bridges with vessels at each side which contained the same solution, and into these were led metallic electrodes with a current of 2-6 volts, and 0.1 to .001 milliampere. Neglecting the effects of stronger currents, which produce curvatures due to death or disturbances of growth, the vast preponderance of response was a turning toward the anode, NH<sub>4</sub>Cl alone showing 6 out of 8 curvatures toward the cathode. Chemotropic studies have shown that the reaction changes with concentration; it likewise

<sup>9</sup> International Catalogue of Scientific Literature. Fourth annual issue. M. Botany. Published for the International Council by the Royal Society of London. London: Harrison & Sons. 37s. 6d.

<sup>10</sup> Semi-annual report of SCHIMMEL & Co. (FRITSCHE BROS.). Miltitz near Leipzig. 12mo. pp. 161. New York: Fritsche Bros. 1906. Free.

<sup>11</sup> DALLA TORRE, C. G. DE, and HARMS, H., Genera Siphonogamarum ad systema Englerianum conscripta. Fasc. 9, pp. 641-720. Leipzig: Wilhelm Englemann. 1907. M 6.

<sup>12</sup> SCHELLENBERG, H. C., Untersuchungen über den Einfluss der Salze auf die Wachstumsrichtung der Wurzeln, zunächst an der Erbsenwurzel. Flora 96:474-500. 1906.

reverses with increasing concentration of solution when an electric current traverses it. Further, the reaction was found to be an additive effect, depending upon the sum of the specific actions of the cathions and the anions. From all the phenomena the author is led to a conclusion, neither novel nor very well founded, that with salts chemotropism and galvanotropism are identical, both being explicable by the migration of ions into the cell and the disturbance of electrical equilibrium they there produce. In chemotropism the migration is due to a concentration gradient, in galvanotropism to the electric current. When non-electrolytes call forth chemotropic responses (as certainly they do) SCHELLENBERG would explain the action by a modification of the permeability of the plasma to the internal salts, whose unequal outward diffusion disturbs the inner electric equilibrium. The experimental work of this paper lacks definiteness and seems to be devoid of sufficient precautions.

GASSNER, working in KNY's laboratory in Berlin, concludes that galvanotropism is only a form of traumotropism.<sup>13</sup> His seedlings were placed in small boxes with perforated cork bottoms, through which the roots protruded into a vessel of tap water, frequently renewed, and through an equal cross-section of this passed the electric current from carbon electrodes of uniform size. He used the commercial current of 110 volts, up to 17 milliamperes. But as he found current density (strength of current divided by its cross-sections) to be a controlling factor, *caeteris paribus*, he states this always in milliamperes per cm<sup>2</sup>. (To this factor SCHELLENBERG seems to have paid no attention.) On that basis the currents used varied from 0.001 ma. to 5 ma., those most used lying below 0.1 ma.

With weakest currents there was no curvature; with currents from a certain density and time of action up, a curvature toward the cathode; with stronger currents or longer time, first a curvature toward the anode in the part above the growing zone, then a paratonic growth-curvature toward the cathode in the growing zone, thus producing S-curvatures; with further increase in current, complete curvature toward the anode, the injury retarding or inhibiting growth on the cathode side even in the growing zone; with still stronger current, partial curvature toward the anode, growth being soon arrested by death; finally, no curvature with currents that cause death too promptly. Differences were observed in the response of different plants to like currents. The positive (toward anode) and negative curvatures are held to be of different nature. The former, earlier in time, is due to injury by reduction of turgor in full-grown cells on the anode side; the latter to retardation of growth on the cathode side of the growing region. (Since a current which will produce only positive curvature in a given time will with longer duration produce negative curvature, it would seem that such a distinction in "nature" could hardly be valid.) The negative curvature is analogous to curvatures due to geotropism, traumotropism, etc., in which the root tip is the percipient part.

<sup>13</sup> GASSNER, G., Der Galvanotropismus der Wurzeln. Bot. Zeit. 64: 149-222. figs. 12. 1906.

All curvatures, especially the S-ones, are parallel with traumotropic curvatures. GASSNER rejects BRUNCHORST's explanation (the action of the products of electrolysis, to which in essence SCHELLENBERG adheres) and that of RISCHAWI (accumulation of water on the convex side), claiming that galvanotropism is only a special case of traumotropicism, in which the injury to the positive side is probably wrought by the passage of the current, as happens with other semipermeable membranes. Admitting that the electric current probably produces its effects by reason of migration of ions, GASSNER definitely declines to identify galvanotropism with chemotropism, since the latter itself may be only a modified form of traumotropicism or indeed of osmotropism. Nor does he think the entry of hydroxyl ions on the anode side can account sufficiently for the injury, because by calculation their amount is infinitely small, and in an experiment rootlets of corn, containing red anthocyan, showed no change of color, though they curved well in a strong current. Rather he would ascribe the injury to the emigration of ions from the plasma.

The observations in these two papers are not so wide apart, nor are the interpretations so antagonistic as they at first appear.—C. R. B.

**Paleozoic botany.**—In his presentation of the present status of paleozoic botany, SCOTT<sup>14</sup> dismisses the lower cryptogams with the brief space (7 pages) which their recorded occurrence in the paleozoic strata warrants, and devotes the remainder of his article to the Vasculares. He adopts provisionally the division of vascular plants into two phyla, the Lycopida and the Pteropsida, as proposed by JEFFREY. Under the Lycopida are ranged the following classes: Sphenophyllales, Equisetales, Psilotales, and Lycopodiales. The first two classes are included under a group name, Articulatae, a propinquity of relationship thus being recognized, which was first pointed out by JEFFREY and subsequently by LIGNIER. In his treatment of the Sphenophyllales the author describes the features of the various types, already for the most part generally known from his textbook. One genus new to the general student is NATHORST's *Pseudobornia* from the Upper Devonian of Bear Island, which is regarded by its author as the type of a special class, the Pseudoborniales. It is characterized by highly dichotomously divided and pinnatifid leaves, which have a certain resemblance to fern-fronds, a resemblance which is considered by SCOTT as of sufficient importance to indicate a certain affinity between the sphenophyllaceous stock and that of the Filicales. Unfortunately nothing is yet known of the internal structure of *Pseudobornia*. The author regards the characters of *Psilotum* and *Tmesipteris* as sufficiently distinct from those of the Lycopodiales to warrant their separation as a special class, the Psilotales. Indeed, he is of the opinion that their peculiar sporophylls find their nearest counterpart in those of the Sphenophyllales. The author even states that if he had to choose between lycopodineous and sphenophyllaceous antecedents for his new class he would choose the latter.

<sup>14</sup> SCOTT, D. H., The present position of paleozoic botany. *Progressus Rei Botanicae*, redigiert von J. P. Lotsy, pp. 139-217. 1906.

Under the Pteropsida are grouped the classes Filicales, Pteridospermeae, and Gymnospermeae, the two latter being regarded as of coordinate importance and constituting the Paleozoic Spermophyta. The author in his discussion of the Filicales makes a strong stand against the tendency to include all paleozoic filicoid plants in the Pteridospermae. He points out that there is clear evidence that the Botryopterideae of RENAULT were true ferns. This is likewise in all probability true of other apparent filicinean remains, of unresolved affinity, characterized by the presence of annulate sporangia, for which the author proposes the form-generic name *Pteridotheca*. There is further good evidence in fruiting fronds referred to Ptychocarpus, Asterotheca, etc., with synangial sporangia resembling those of existing Marattiaceae, as well as in stems presenting the anatomical structure of Psaronius, for the existence of ferns like the Marattiaceae in paleozoic times.

The paleozoic seed-plants are included under two coordinate classes: the Pteridosperm(e)ae and Gymnosperm(e)ae, which are spelled with an unusual and perhaps superfluous *e*. The description of the Pteridospermae (Cycadofilices of POTONIÉ) contains little which is not to be found in the author's lecture before the Vienna Congress. He states very clearly however his reasons for regarding the Pteridospermae as a group coordinate with the remaining gymnosperms as at present recognized: (1) the mega- and microsporangiate sporophylls were little modified from ordinary vegetative fronds; (2) the anatomical structure was more clearly fern-like than that found in any other gymnosperms. It may well be objected, however, that the Pteridospermae were essentially gymnosperms, that the Cycadophyta, taken as a group, present equally fern-like mega- and microsporophylls, and that the anatomical peculiarities of the Pteridospermae can nearly all be duplicated in the lower Gymnospermae. Whether or not the Pteridospermae stand as a distinct class coordinate with the remaining Gymnospermae, there can be no question that their discovery constitutes the principal advance in many years in our knowledge of the Spermaphyta. All botanists must be grateful to the author for his lucid and interesting account of paleozoic plants, which he has done so much to restore and rescue from oblivion. In looking over these pages, one is tempted to call the article an original compilation, so large and important, albeit not in any way disproportionate, a part, do the author's own investigations make of the whole.—E. C. JEFFREY.

**Cytological studies on the Cyanophyceae.**—Two contributions<sup>15, 16</sup> on the cytology of the Cyanophyceae have recently appeared, which add a few new features to an already much confused subject. GARDNER regards as the chief trouble which has obscured the truth for former investigators their "failure to discover a method which would clearly, definitely, and unmistakably differen-

<sup>15</sup> GARDNER, N. L., Cytological studies in Cyanophyceae. Univ. Calif. Pub. Bot. 2:237-96. pls. 21-26. 1906.

<sup>16</sup> GUILLERMOND, A., Contribution à l'étude cytologique des Cyanophycées. Rev. Gén. Bot. 18:392-408, 447-65. pls. 9-13. 1906.

tiate the structures which are present." He has discovered that material treated with a strong solution of iodin; then washed with 95 per cent. alcohol; finally placed in water and pressed upon and rolled under a cover glass gives preparations "in many ways superior to microtome sections." Hence the many colored figures which accompany his paper are all drawn from optical cross or longitudinal sections rather than from microtome sections. Such a method appears to the reviewer entirely inadequate to solve the difficult problems pertaining to the cells of the Cyanophyceae.

The central body, which is regarded by GARDNER as a nucleus, consists of a thread-like structure composed of chromatin (as evidenced by staining reactions); a ground substance (not differentiated in his drawings); and " $\alpha$ -granules" (the slime globules of former authors). The ground substance acts, in his opinion, simply as a matrix for the chromatin elements and plays no essential rôle in nuclear division; as to whether this is achromatin or not, the author withholds judgment. Three types of nuclei are distinguished: (1) the diffuse type, which is characteristic of most of the fifty forms examined; (2) the net-karyosome type, found only in *Dermocarpa*—which presents a new kind of nuclear division, in that a large nucleus is represented as breaking up simultaneously into a large number of daughter nuclei; and (3) the primitive mitosis type, found likewise in one species only. The nuclei of types 1 and 2 divide by simple amitosis; while in type 3 the breaking up of the chromatic thread into three long parallel segments is regarded as a step toward mitosis.

GARDNER finds no special chromatophore. The first product of assimilation, in his opinion, is grape sugar, some of which is converted into glycogen and thus stored. He adds some interesting experiments on the effects of various changes of habitat, such as from salt water to fresh and to distilled water, alternate drying and submergence in water, the effect of prolonged darkness, etc. He finds that such changes do not produce any marked change in cytological characters.

GUILLIERMOND, who also apparently used no sections in his study, is in general agreement with GARDNER's conclusions. He prefers, however, to regard the central body as a sort of nucleus reduced to the state of a "chromidial" network. It is in reality "un noyau sans membrane" (a conclusion which nearly all other investigators who accept the nuclear nature of the central body have held before him). GUILLIERMOND comes to one conclusion, however, which is apparently new, namely that there are present in the central body two kinds of granules, whereas GARDNER as well as most other investigators have distinguished only one. The one or two large, conspicuous granules of the central body GUILLIERMOND refers to the nucleolus-like bodies of A. MEYER (in my opinion the so-called slime globules). Besides these, he distinguishes the metachromatic granules; smaller numerous granules which he regards as corresponding to the slime globules (PALLA), or red granules (BÜTSCHLI), or anabaenin granules (FISCHER), or volutin granules (MEYER). Their appearance in some of his drawings suggests, however, what have been called by the reviewer, in a paper on the subject, chromatin granules. It seems likely, in fact, that Guill-

LIERMOND, at least in some of his drawings, has combined in his metachromatic bodies two distinct kinds of granulations, namely, chromatin granules and slime globules.—EDGAR W. OLIVE.

**Papers on algae.**—The following papers on algae appear in the volume of papers issued by the students of Dr. F. R. KJELLMAN in honor of his sixtieth birthday.

BORGE<sup>17</sup> lists forty-six species (mostly desmids) of fresh-water Chlorophyceae, of which several are new to science, from collections made by Dr. P. DUSÉN in Magellan Ter. in 1896, and in Tierra del Fuego Ter. and Desolation Island in 1906. He also enumerates 77 species, comprising all the fresh-water Chlorophyceae known at the present time from the islands south of Magellan Strait.

KYLIN<sup>18</sup> describes and figures four species of Chantransia, three of which are new to science.

SVEDELIUS<sup>19</sup> made a thorough study of the Ceylon reef vegetation at Point de Galle in the year 1902-03; first at the time of the winter or northeast monsoon (November-March) and later at the time of the summer or southwest monsoon (August). His observations were carried on with a view to establishing the existence of a periodicity in the marine vegetation of this region, and to showing how this was manifested and from what it resulted. He found that there is an undoubted antagonism between living coral and algal growth. Only certain calcareous algae and forms with creeping rhizomes, such as *Caulerpa clavifera*, can survive. Algal life was richest upon dead coral ledges and exposed rocks. The Florideae unquestionably dominate the vegetation of the littoral region at Point de Galle. The color of these forms is peculiar, being dark violet, gray-brown, or gray-green, instead of red or purple. It was proved that a marked periodicity prevails in the littoral algal flora on the coast of Ceylon. Certain species occur only at a certain time during the year, and their period of development is extremely limited. The chief mass of vegetation is made up of perennial species which live throughout the year; of these many produce fruit only during a certain time. Some species show no indication of periodicity, but appear the same at all times of the year. It was established that the periodicity depends upon the change of monsoon (*Monsunwechsel*), but the exact causes for this are not yet made clear. Apparently the condition of the water (temperature, salinity, amount of agitation), as influenced by the monsoon, might have considerable effect upon the periodicity.

SKOTTSBERG<sup>20</sup> accompanied the Swedish Antarctic Expedition and was in

<sup>17</sup> BORGE, O., Süsswasser-Chlorophyceen von Feuerland und Isla Desolacion. *Botaniska Studier, tillägnade F. R. KJELLMAN*, 21-34. *pl. 2.* *figs. 5.*

<sup>18</sup> KYLIN, HARALD, Zur Kenntnis einiger schwedischen Chantransia-Arten. *Op. cit.* 113-26. *figs. 9.*

<sup>19</sup> SVEDELIUS, NILS, Ueber die Algenvegetation eines ceylonischen Korallenriffes mit besonderer Rücksicht auf ihre Periodizität. *Op. cit.* 184-221. *pl. 6.* *figs. 10.*

<sup>20</sup> SKOTTSBERG, CARL, Observations on the vegetation of the antarctic sea. *Op. cit.* 245-64. *pls. 7-9.*

antarctic regions during January and February 1902, and in November and December of the same year. The South Shetland Islands and Graham Land (Louis Phillippe Peninsula, the Palmer Archipelago, Ross Island, and Snow Hill Island) were visited. The purpose of the paper is to give a short description of the meteorological and hydrographical conditions which rule the antarctic marine flora, a general survey of its appearance, and a rough sketch of its systematic composition. A list of 35 species of antarctic algae is furnished. According to the author, "the general character of the flora of the Graham region discloses . . . several interesting, probably ancient, endemic types, and a large percentage of subantarctic, especially magellanian, species. Nothing favors the so-called bipolar forms."—JOSEPHINE E. TILDEN.

**Morphology of *Cephalotaxus drupacea*.**—It is interesting to compare the results of the study of *Cephalotaxus Fortunei* by COKER<sup>21</sup> with the results obtained by LAWSON<sup>22</sup> in a study of *C. drupacea* and published almost simultaneously. The two accounts are so nearly identical in the main features, that one may have a feeling of confidence that our knowledge of the structures described in these papers is well established for the genus. *Cephalotaxus* has been a genus of special interest on account of its supposed primitive character, emphasized in recent years by the study of its vascular anatomy. The studies of the reproductive structures seem to contradict this claim to a certain extent, as have all recent similar studies of other Taxineae.

The main results of LAWSON's study of *C. drupacea* may be summarized as follows: In the germination of the microspore no prothallial cell is cut off, and before pollination the generative and tube nuclei have been organized. Pollination occurs late in March, but no further nuclear divisions take place until the following spring, when the pollen tube begins to penetrate the nucellus and the generative nucleus divides. When the tube has reached the archegonial chamber, which takes about ten days, the nucleus of the body cell divides, forming two sperm nuclei of equal size, but with no wall-formation. The female gametophyte develops in the usual way, with free nuclear divisions, vacuolation and the parietal placing of the free nuclei, wall-formation, and centripetal growth. Four archegonia are organized, each with a distinct chamber, and with two or frequently three neck cells. A ventral canal nucleus is cut off and degenerates before fertilization. In fertilization the entire contents of the pollen tube are discharged into the egg, but the two sperm nuclei are not released from the membrane of the body cell until the interior of the archegonium is reached. The fusion nucleus gives rise to four free nuclei near the center of the egg, which pass toward the bottom, accompanied by the various food materials, where divisions continue until sixteen free nuclei are formed, when the first walls appear. Finally four

<sup>21</sup> COKER, W. C., Fertilization and embryogeny in *Cephalotaxus Fortunei*. BOT. GAZETTE 43:1-10. pl. I. figs. 5. 1907.

<sup>22</sup> LAWSON, A. A., The gametophytes, fertilization, and embryo of *Cephalotaxus drupacea*. Annals of Botany 21:1-23. pls. I-4. 1907.

tiers of walled cells are organized, the uppermost being the "rosette," the next developing the suspensor, the third forming the embryo, and the terminal one developing the "penetrating cap" characteristic of the genus. The embryo may reach the 16- or 32-celled stage before the suspensor begins to elongate. After the full development of the suspensor a series of long "embryonal tubes" arise from the proximal cells of the embryo. An interesting observation is the budding out of small secondary embryos from the main group of embryo cells, though ordinarily but a single embryo is produced by the fertilized egg.—J. M. C.

**Coastal plain of Georgia.**—Concerning none of the older settled portions of the United States has there been so much difficulty in obtaining accurate information on plant distribution as for the extreme southeastern coastal plain. Because of its recent origin as a land area, its proximity to one of the most ancient land masses, and its connection with the tropics by way of the Florida peninsula, it forms one of the most critical regions on the continent for the investigation of plant origin, migration, and acclimatization. R. M. HARPER<sup>23</sup> has recently published the results of his investigations in southern Georgia and has made an important contribution to North American phytogeography. The classification of his observations, the clearness with which the results are presented, and the excellence of the illustrations add much to the value of the paper.

The first part of the volume is devoted to a brief summary of the geological divisions of eastern North America and the subdivisions of the Georgia coastal plain. The Altamaha Grit forms a strongly marked physiographic region. It is probably Pliocene in age, occupies the middle third of the coastal plain of Georgia, and its topography is typically rolling. Rock outcrops are rare, the soil being formed mostly by the overlying LaFayette (sand and clay) and Columbia (sand) formations. The vegetation is discussed under nineteen "habitat groups," among which are rock outcrops, dry pine-barrens, moist pine-barrens, swamps, cypress ponds, sand hills, and hammocks. In each case the plant list is most carefully analyzed and shows at a glance the trees, shrubs, vascular and non-vascular herbs, their relative abundance, duration, flower color, and evergreen or deciduous habit. Each list is accompanied by a phenological diagram exhibiting the times of flowering. The accompanying descriptions give the characteristics of the habitat, ecological notes, geographic ranges of the plants, and their taxonomic relationships. In the final summary the relations of the typical habitat groups to each other and to other regions are represented by diagrams; some exceptional habitats are described, the weeds are listed, and the effects of civilization discussed.

The second part of the paper presents a history of botanical exploration in the region, an annotated catalogue of the species, a summary of the catalogue, and a list of the papers consulted.—E. N. TRANSEAU.

<sup>23</sup> HARPER, ROLAND M., A phytogeographical sketch of the Altamaha Grit region of the coastal plain of Georgia. *Annals N. Y. Acad. Sci.* 17:1-415. *pls. 1-28.* 1906.

**Constriction of twigs by bag-worm.**—VON SCHRENK publishes an account of interesting observations made by him on the constriction and consequent deformation or death of twigs, produced by the bands, about 3<sup>mm</sup> wide, spun by the bag-worm (*Thyridopteryx ephemeraeformis* Haworth) for suspending its cocoon in late summer from the slender twigs, 3<sup>mm</sup> in diameter or thereabouts, of various deciduous and evergreen trees. Most of these bands are burst by the renewed growth of the twig in the second year of its age and the "bag" drops off; but 1.5-2 per cent. of the bands are too strong, and by resisting growth cause a deformity in the region of the girdle. VON SCHRENK has studied the anatomical changes produced, and the pressure to which, by its own growth, the twig is subjected. This he believes is two or three times the 15 atmospheres given by KRABBE as the pressure under which growth may continue. VON SCHRENK, however, bases his conclusions upon the breaking strength of the band, which indicated in tests of 400 a possible radial pressure of 4 to 162 atmospheres, the majority running from 14 to 44. The tested bands, however, were those not broken by the twig, and it is quite possible that the pressure due to growth reaches a maximum which was insufficient to rupture the band (perhaps far too little), but adequate to stop further growth under it.

The argument that the excessive growth on one or both sides of the girdle is due to the fact that "the band has evidently stopped the passage of plastic materials through the bark absolutely," or partly, or temporarily, and "as a result of this an accumulation of these substances has taken place and the cambium layer . . . has formed . . . cells to an enormous degree, as a result of which the outer part of the twig has grown very much in diameter," is one that seems to put the cart before the horse. Is it not rather that this pressure acts as a stimulus and thus accelerates the growth of near-by cells, which itself brings to this place an adequate supply of food?—C. R. B.

**Chemotaxis.**—KNIEP, working in PFEFFER's laboratory and using his well-known capillary-tube method, has sought to determine<sup>24</sup> whether various factors may strengthen or weaken or even abolish the sensitiveness of bacteria to definite reagents; and whether the variation in sensitiveness under such conditions holds for all chemical stimuli or is limited to certain ones, sensitiveness remaining unaltered toward others or being affected only by other conditions. He finds that definite alterations in the external conditions do excite or hinder at pleasure the reaction of certain bacteria to one group of substances, while the reaction to another is not affected. And this shows, he argues, that we have to do not merely with a chemical preceptivity, but with several; how many, is not known, for it is quite conceivable that different irritabilities possess a common property of being called out or released by the same change in the external conditions.

The changes thus effected in the sensitiveness of the bacteria may be themselves responses; and they may act by altering the intensity of stimulation or

<sup>24</sup> KNIPE, HANS, Untersuchungen über die Chemotaxis von Bakterien. Jahrb. Wiss. Bot. 43:215-270. 1906.

the character of the reaction, or by altering both. Cases of this kind are cited from his own researches and those of KLEBS, ROTHERT, VÖCHTING, and others. This variation in sensitiveness has a parallel in that of other physiological characters, as, for example, the behavior of aerobes toward oxygen, luminescence, pigment-formation, etc. The specific effect of acids and alkalies is likened to the effects of  $H^+$  and  $OH^-$  ions on various functions. Other observers, notably SHIBATA, have shown similar chemotactic sensitiveness of spermatozooids to both anions and cathions.

The author points out "interesting and far-reaching analogies" between the *chemischer Sinn* of bacteria and the corresponding *Geschmackssinn* of man, and suggests that further investigations may find these phenomena more complicated than they now appear.—C. R. B.

**Camptotropism and geotropism.**—Two new terms are added to the vocabulary of irritable phenomena, and some interesting reactions described by BÜCHER.<sup>25</sup> It appears that when a shoot capable of growth is forcibly bent, it reacts to the tensions thereby set up by thickening the walls and reducing the cell diameter in the mechanical tissues (collenchyma, bast, wood) on the convex side, and conversely, forming thinner walls and larger cells on the concave side. This reaction is due to *Kamptotropismus*, being interpreted as belonging to the same category as WIESNER's heterotrophisms. In like manner when a similar shoot, geotropically sensitive, is fastened in the horizontal position and prevented from responding to gravity, anatomical changes of the same kind ensue, which BÜCHER ascribes to geotropism. It is unfortunate that the new term is so like the well-established geotropism, with which it will be difficult to prevent confusion, even in English speech.

Simple strain, in the normal position, as BALL showed, does not effect such anatomical changes; but both pressure and traction are effective in the phenomena referred to. The presentation time varies from 24 to 48 hours and the reaction time in Ricinus and Phaseolus is about 3 days. Forceful curvature and restrained geotropic response combined give a summation effect; opposed, one usually prevails strikingly over the other. In Ricinus the geotropic, in Abutilon and Euphorbia the camptotrophic response dominates. A similar reaction was found in heliotropic stems; analogously, heliotropism may be predicated. In the excentric growth of the lateral branches of trees there are special reactions which affect the activity of the cambium and its differentiation, but geotropism is restricted to young parts. Various observers have shown that there are also other causes for heterotropism.—C. R. B.

**Life-history of cotton.**—BALLS<sup>26</sup> has studied the life-history of *Gossypium* from floral development to the early stages of the embryo. Aborted anthers are

<sup>25</sup> BÜCHER, H., Anatomische Veränderungen bei gewaltsamer Krümmung und geotropischer Induktion. *Jahrb. Wiss. Bot.* 43:271-360. figs. 40. 1906.

<sup>26</sup> BALLS, W. L., The sexuality of cotton. Reprint from *Yearbook of the Khedivial Agric. Soc.* Cairo. 1905. pp. 26. pls. 9.

common, the failure occurring either directly after synapsis, the chromatin thread not being segmented into chromosomes, or after tetrad formation, the spores aborting. The microsporangiate archesporium consists of a hypodermal plate of cells (two cells in transverse section and six in longitudinal). In connection with the reduction division, 20 chromosomes were counted as the gametophyte number; and this was checked up by an approximate count of 40 in certain nuclear divisions of the embryo. The solitary megasporangiate mother cell is differentiated beneath a heavy development of nucellar tissue (about 12 layers). One of the surprising results is that the functioning megasporangiate of the linear tetrad is the micropylar one. The antipodal cells are evanescent, and the fusion of the polar nuclei is somewhat tardy. Double fertilization was observed very distinctly. In the development of the embryo no suspensor was discovered, and before the segmentation of the fertilized egg about 100 free, parietally placed, endosperm nuclei have appeared. Later there is endosperm wall-formation, and in the later stages of the embryo it is imbedded in a delicate endosperm tissue. Perhaps the most interesting data are those in reference to the time-relations of these events, the rate of development being unusually high. Flowers hand-pollinated at 10:00 A. M. showed fertilized eggs in the afternoon of the following day. The interval between megasporangiate-formation and the completed sac (fertilization stage) is three days. About 60 hours after fertilization ( $3\frac{1}{2}$  days after pollination) the egg segments, and in about a week after pollination the embryo consists of "hundreds of cells."—J. M. C.

**Pteridosperms and angiosperms.**—OLIVER has published the substance of a lecture delivered before the Botanical Club of Cambridge University<sup>27</sup> and illustrated it by a scheme of the occurrence of vascular plants in geological time, modified from E. W. BERRY. It discusses the bearing of recent investigations of the pteridosperms and cycads on the origin of seed-plants. Attention is called to the fact that the appearance of the Cycadophyta is geologically synchronous with the disappearance of the pteridosperms, and that the former perpetuate in many respects, especially in the cycadeoidean forms recently described by WIELAND, the fern-like characters of the latter. Just as the incoming of the Cycadophyta marks the end of the reign of the pteridospermic gymnosperms, so the appearance of the angiosperms in the Cretaceous and Tertiary marks the waning of the Cycadophyta. The author calls attention to the angiospermoid protection of the exalbuminous seeds of Cycadeoidea by the sterile scales of the inflorescence. He further emphasizes the resemblance of the bisporangiate inflorescence with its perianth-like envelope in Cycadeoidea with the typical angiospermous flower, since the parts occur in the same order, namely, perianth, microsporophylls, and, uppermost of all, the megasporophylls. He suggests that possibly the Cycadophyta may be appropriately divided into two series, the Gymnocyads and the Angiocycads, the former the ancestors of the living cycads and the latter consti-

<sup>27</sup> OLIVER, F. W., Pteridosperms and angiosperms. *New Phytologist* 5:232-242. 1906.

tuting the Bennettitales. The latter or some allied stock he regards as the possible ancestors of the angiosperms, a probability which he considers strengthened by the incoming of the angiosperms as they became extinct.—E. C. JEFFREY.

**Stem-thickening in monocotyledons.**—It has been claimed that in palms secondary thickening by means of a merismatic zone occurs, as in *Dracaena*. STRASBURGER<sup>28</sup> has investigated the subject by a study of two stems of *Washingtonia filifera*, and finds no cambial zone, but instead a number of localized areas in the pericycle where fundamental tissue, bundles, and sclerenchyma are produced. An examination of stems of *Pandanus utilis* afforded similar results. Among the numerous details recorded is the observation concerning the arrangement of the leaves in *Pandanus*; following SCHWENDENER the author finds that this is due to torsion of the stem axis.

Another paper on Pandanaceae, by CARANO,<sup>29</sup> denies the existence of secondary growth in this family. Among other observations of interest is the crushing of vessels of the xylem at the leaf bases by growth of the wood parenchyma; this process seems to aid in casting off the old leaves. The bundles of the blade of the leaf show a horseshoe-shaped mass of phloem surrounding the xylem, and water-storage tissue is abundant along the median nerve. The root shows a number of large medullary bundles in addition to the peripheral ones, and to the former the lateral roots are attached. The roots springing from the stem are likewise connected with deeply situated vascular strands. These features hardly support the claim that this family is a primitive one.

In the palm *Euterpe oleracea*<sup>30</sup> the stem increases in thickness by the extraordinary growth of the mechanical tissue lying external to the phloem. This growth takes place after the stem has become hard and woody.—M. A. CHRYSLER.

**Germination of dimorphic fruits.**—ERNST<sup>31</sup> has investigated the germinative characters of the dimorphic fruits of *Synedrella nodiflora*, a composite of the East Indian Archipelago. He finds that the fruits produced by the pistillate-ray flowers differ radically in form from those produced by the perfect disk flowers. Corresponding to these differences in structure, he finds differences in germinative characters. The disk fruits germinate both in light and in darkness to a higher per cent. and more quickly than do the ray fruits. In the disk fruits diminution in the intensity of the light or its complete withdrawal only retards slightly the first stages in the germination, although it defers considerably the

<sup>28</sup> STRASBURGER, E., Ueber die Verdickungsweise der Stämme von Palmen und Schraubenbäumen. Jahrb. Wiss. Bot. 43:580-628. pls. 3-5. 1906.

<sup>29</sup> CARANO, E., Ricerche sulla morfologia delle Pandanacee. Annali di Botan. 5:1-46. pls. 1-5. 1906.

<sup>30</sup> KRÄNZLIN, H., Ueber das Dickenwachstum der Palme *Euterpe oleracea*. Ber. Deutsch. Bot. Gesells. 24:483-489. 1906.

<sup>31</sup> ERNST, A., Das Keimen der dimorphen Früchten von *Synedrella nodiflora*. Ber. Deutsch. Bot. Gesells. 24:450-458. 1906.

development of the hypocotyl and the unfolding of the cotyledons. In the ray fruits diminution of the intensity of the light and its complete withdrawal cut down markedly both the speed and per cent. of the first steps of germination; while in darkness the germination never goes beyond the early stages. The favorable influence of white light on the early germination and the later development of the seedling of the ray fruits is due to the less refrangible rays. The strongly refrangible rays retard the early stages of germination of the ray fruits and inhibit the further development of the seedlings. While ERNST holds that the differences in the germinative characters of the two fruits are due to differences existing in the living portions of the fruits, it is possible, in the light of the results of another investigation,<sup>32</sup> that they lie in the coats, and that his results with light of different intensity and refrangibility came from accompanying variations in temperature, of which apparently he kept no record.—WM. CROCKER.

**A new type of Medullosoeae.**—The material described by SCOTT<sup>33</sup> was derived from an abandoned coal mine at Shore-Littleborough in Lancashire, which was opened for scientific purposes by its owner Mr. W. H. SUTCLIFFE, in honor of whom the new genus is named. It presents a strong general resemblance to *Medullosa*, but differs strikingly from it in the fact that it possesses a single axial protostele instead of the numerous concentric strands which are found in the latter genus. Another noteworthy feature is the exarch character of the wood, in contrast to the mesarch condition found in other Medullosoeae. In spite of the simple character of the caudine vascular system, the leaf-strands are numerous and undergo complex branching and anastomosis. The foliar bundles are concentric. Secondary thickening is present to a very slight degree in the central cylinder of the stem. The complex leaf supply attached to a protostelic caudine cylinder is a unique condition, and, as the author has pointed out, may have resulted from a reduction of the central cylinder of the stem. This view may perhaps also be taken of the cambium, which is more likely to be vestigial than inchoative. Mucilage canals and reticulate peripheral bands of sclerenchyma are found to resemble those found in *Medullosa*. The photographic illustrations by Mr. L. A. BOODLE are of quite unusual beauty.—E. C. JEFFREY.

**Items of taxonomic interest.**—W. B. HEMSLEY (*Hooker's Icônes Plant.* IV. 9:1906) has described the following 3 new genera from the Seychelles: *Indokingia* (*pl. 2805*) and *Geopanax* (*pl. 2821*) belonging to Araliaceae, and *Neoschinamera* (*pl. 2810*) belonging to Rubiaceae; and from China a new genus (*Sinowilsonia*, *pl. 2817*) of Hamamelidaceae.—O. STAPP (*idem*) has described a new genus (*Elaeophorbia*, *pl. 2823*) of Euphorbiaceae from western Africa.—W. H. BLANCHARD (*Rhodora* 9:4-10. 1907) has described 3 new species of *Rubus* from Connecticut.—E. HASSSLER (*Bull. Herb. Boiss.* II. 7:1-4. *fig. 1.* 1907) has

<sup>32</sup> CROCKER, WM., Rôle of seed-coats in delayed germination. *Bot. GAZETTE* 42:265-291. 1906.

<sup>33</sup> SCOTT, D. H., On *Sutcliffia insignis*, a new type of Medullosoeae from the Lower Coal Measures. *Trans. Linn. Soc. London* II. 7:45-68. *pls. 4.* 1906.

described a new genus (*Pseudomachaerium*) of Leguminosae (Dalbergieae) from Paraguay.—P. B. KENNEDY (Muhlenbergia 3:8. 1907) has described a new Trifolium from Montana.—T. D. A. COCKERELL (*idem* 9) has published a new genus (*Microbahia*) founded on *Syntrichopappus Lemmoni*.—A. A. HELLER (*idem* 10-12) has described new western species under Cakile and Ribes.—A. NELSON and P. B. KENNEDY (Proc. Biol. Soc. Wash. 19:35-40, 155-158. 1906) have described new species from Nevada under Eriogonum (2), Arabis, Ribes, Gilia, Phlox, Castilleja, Hulsea, Raillardella, Chrysanthmus, Sophia, Sphaerostigma, Godetia, Oreocarya, Cryptantha (3).—J. M. C.

**Ecology and taxonomy of Caulerpa.**—SVEDELIUS<sup>34</sup> has published the results of his studies of the Ceylonese species of Caulerpa. In a discussion of their mode of life the following subjects are presented: (1) "do all Caulerpas grow under similar external conditions?"; (2) "different ecological types as distinguished by the varying development of their root-system," and (3) "of their assimilation system;" (4) "on the difference between morphological and adaptational characters in Caulerpas." Observations of the different kinds of variations result in the following enumeration: (1) those which depend on locality, and which are to be considered adaptations or "ecologisms;" (2) those which cannot be considered as ecologisms, but are the result of fluctuating variability among the different branchlets; (3) those which can be considered as phylogenetic stages of evolution; (4) bud variations of atavistic origin; (5) dwarf forms; (6) those which do not fall under any of these categories and may be bud variations without atavistic origin (=mutations).

In the taxonomic part of the paper twenty-one species are presented and illustrated, two of them being new.—J. M. C.

**Experiments on cell-formation.**—By subjecting root tips to various abnormal factors, such as  $MgSO_4$ ,  $NaCl$ , benzine vapor, and extreme temperatures, NĚMEC<sup>35</sup> finds that cell walls do not always appear between the daughter nuclei and that the daughter nuclei frequently fuse, the resulting nucleus of course having the double number of chromosomes. In the tips subjected to benzine vapor, the chromosomes are shorter and thicker than the normal ones, although in their formative stages no abnormalities can be observed. He was most successful with root tips of *Vicia Faba*, *Galtonia candicans*, *Hyacinthus*, and various euphorbias. Stamine branches of *Larix decidua* were placed in a warm chamber and subjected to the action of chloroform vapor: There was little doubt that some pollen mother cells passed directly over into pollen grains. Mixed among these pollen grains true tetrads were occasionally present. Sometimes the pollen mother cells divided once, giving rise to two pollen grains.—W. J. G. LAND.

<sup>34</sup> SVEDELIUS, NILS, Ecological and systematic studies of the Ceylon species of Caulerpa. Ceylon marine biological reports no. 4. pp. 81-144. figs. 51. 1906.

<sup>35</sup> NĚMEC, B., Ueber die Bedeutung der Chromosomenzahl. Bull. Internat. Acad. Sci. Bohême 10: pp. 4. 1906.

## NEWS

PROFESSOR GEORG KLEBS, of Halle, has been called to the University of Heidelberg to succeed the late Professor PFITZER.

DR. WILLIAM TRELEASE, Missouri Botanical Garden, is spending February and March in a botanical expedition in the West Indies.

THE LICHEN COLLECTION of H. E. HASSE, of California, consisting of about 3000 species and many duplicates, has been presented to the New York Garden by Mr. JOHN I. KANE.

PROFESSOR ROLAND THAXTER, Harvard University, has been selected as American editor of the *Annals of Botany*, to succeed Professor W. G. FARLOW, who has withdrawn from the position.

B. E. FERNOW, formerly director of the New York State College of Forestry at Cornell University, has been appointed to organize a department of forestry in the State College of Pennsylvania, which will be an undergraduate forestry school.

MUHLENBERGIA, with A. A. HELLER as editor and publisher, begins its third volume as a journal "open to the botanical public." It is to be restricted to the taxonomy of seed-plants. The subscription price is \$1.00 a year, and the address of the editor is Box 58, Los Gatos, California.

WITH THE January number the *Bryologist*, under the editorship of the publisher, Mrs. ANNIE MORRILL SMITH, enters upon its tenth volume, which opens with a sketch and portrait of the late WILLIAM MITTEN. The past volumes are replete with notes on the bryophytes of the United States.

THE OFFICERS of the American Society of Naturalists for the present year are President, J. P. McMURRICH, University of Michigan; Vice-President, D. P. PENHALLOW, McGill University; Treasurer, HERMANN VON SCHRENK, Missouri Botanical Garden; Secretary, E. L. THORNDIKE, Columbia University.

ADVANCE PROOFS of the *Report* of the Missouri Botanical Garden for 1906 show a very vigorous development in every department. The gardening for the benefit of the public was responded to by 117,553 visitors, a figure not reached heretofore except during the year of the Louisiana Purchase Exposition. To the herbarium there were added 35,422 sheets of specimens, bringing the total collection to 559,267 sheets, 46,466 of which are thallophytes. One of the notable collections added during the year was that of the late J. REVERCHON, especially rich in Texan plants. The library is one of the very few great botanical libraries of America, the accessions of 1906 bringing the enumeration of books and pamphlets to 54,895, controlled by 571,253 index cards. During the latter part of the year J. W. BLANKINSHIP was added to the herbarium staff.

# BOTANICAL GAZETTE

APRIL 1907

## THE FORMATION OF INTUMESCENCES ON POTATO PLANTS<sup>1</sup>

GERTRUDE E. DOUGLAS

(WITH NINE FIGURES)

During the winter of 1904-5, in some experiments to determine the relation of a soil fungus to potato plants, Professor ATKINSON planted some potatoes in pots, and after the young shoots were well started they were covered with bell jars to produce a moist atmosphere, which would be more favorable for the growth of the fungus. In a few days intumescences were formed in great numbers, so that a large part of the plant was literally covered with them. This suggested that the potato plant would be an excellent subject with which to experiment to determine the cause of the formation of these intumescences, at least under certain conditions. The matter was then placed in my hands for this purpose. The subject was the more interesting because of the difference of opinion among plant physiologists as to the influence of light stimulus in the formation of intumescences upon plants, as pointed out by VON SCHRENK (9) in his paper on "Intumescences formed as the result of chemical stimulation."

The methods used in the experiments for producing the intumescences were in general as follows: young plants were covered with bell jars and their roots supplied every day with an abundance of warm water, except in those experiments where the roots were kept cold. The absorption of water was thus very great, while the saturated air under the bell jar greatly checked the transpiration.

<sup>1</sup> Contribution No. 115 from the Department of Botany of Cornell University.

For the purpose of experiment, the light and temperature conditions could be easily changed.

The intumescences were formed very quickly, generally within two to five days after covering the plants. Usually they appeared on the upper side of the leaf, but were observed also, in cases where they were very abundant, on the under side and scattered profusely over the upper part of the stem. They did not form on the very young growing leaves at the tip of the stem and rarely on the two mature lower leaves of the shoot. The appearance of these intumescences was very similar to that described by VON SCHRENK (9) on the cauliflower. To the naked eye they first appeared as very small greenish-yellow dots, projecting slightly from the surface of the leaf. These rapidly developed in size, became lighter yellow, hemispherical, smooth, and glossy. After about twenty-four hours they became whitish and roughened, and projected prominently from the surface. Usually the central part of the intumescence was slightly yellowish in color, with a ring of more whitish cells around the outside. When they first appeared, the intumescences came out over or near the main veins; but when very numerous, they broke out all over the surface of the leaf and looked very much like incrustations of some crystalline salt. The affected leaves curled over toward the under side, and this curling became the stronger the more intumescences were produced. The single intumescences were from 2 to 3<sup>mm</sup> in diameter. When formed numerously, however, the single intumescences often became confluent, making large rough patches. These intumescences lasted but a short time; in a day or two they had collapsed and become dry and blackened. A cross-section through the leaf showed that the intumescences were due to the hypertrophy of the cells lying underneath the epidermis. The swollen cells were first found in the palisade layer; they elongated and pushed against the epidermis; and in most of the cells cross-walls were formed. These cells continued to enlarge until the pressure upon the epidermis caused it to break and the palisade cells to push up through. As the cells enlarged, the chlorophyll granules lost their green color, became yellowish, and disappeared entirely from the cells or remained very much reduced, scattered through the bottom parts.

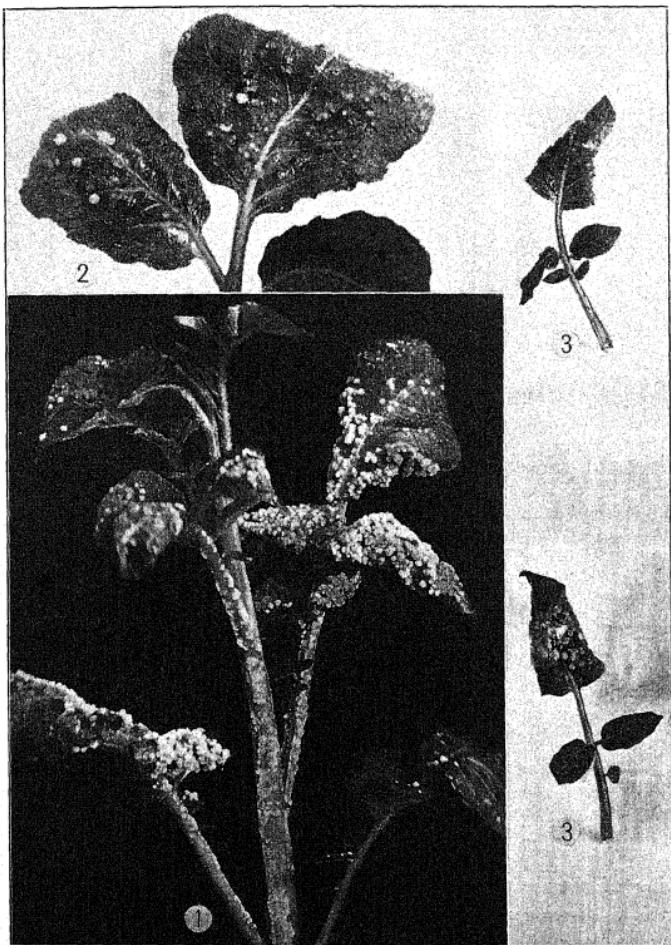


FIG. 1.—Photograph showing intumescences formed on a young potato shoot under whitewashed glass. FIGS. 2, 3.—Intumescences formed on a leaf in sunlight.

After the elongated palisade cells had broken through the epidermis, the pressure against their side walls being thus somewhat relieved, the sac-like cells bulged out laterally and became club-shaped at the tip. The central cells stood up perpendicularly to the surface of the leaf, but the cells around the edge of the intumescences curled over toward the leaf surface, thus causing the outside whitish

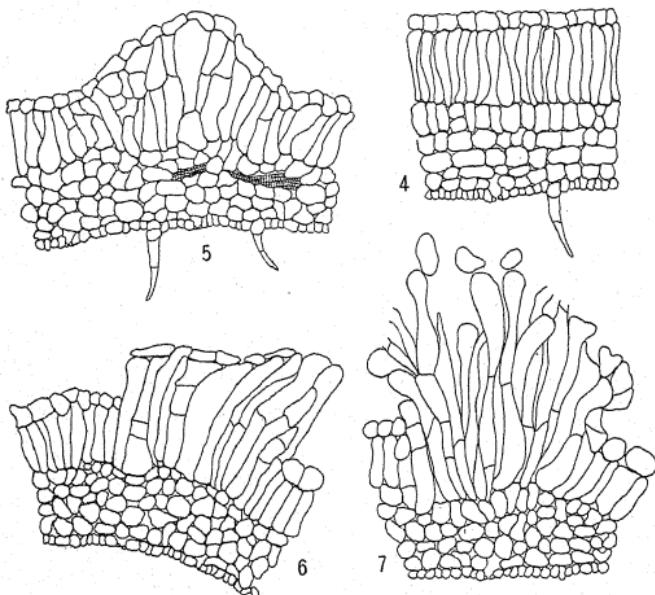


FIG. 4.—Cross-section through a normal potato leaf. FIG. 5.—Cross-section through a young intumescence, showing hypertrophy of palisade layer and formation of cross-walls. FIG. 6.—Later stage; the swollen cells have broken through the epidermis. FIG. 7.—Later stage; some of the cells were cut through in sectioning.

ring of the intumescence, mentioned before. The hypertrophied cells were not confined to the palisade layer, but often included cells in the layer of spongy parenchyma below. In the largest intumescences the swollen cells extended back to one or two rows from the under epidermis. As the intumescences grew old their walls became cutinized.

Intumescences have been observed on various plants by several writers, and all practically agree that they are formed under conditions of excessive humidity accompanied by great warmth. As to

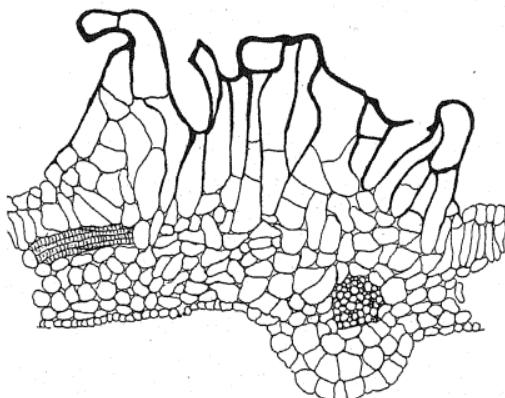


FIG. 8.—Old intumescence; parts of the cells have broken away and the walls have become cutinized.

the presence or absence of a light stimulus, there is a greater difference of opinion. SORAUER (10-20), who has described the majority

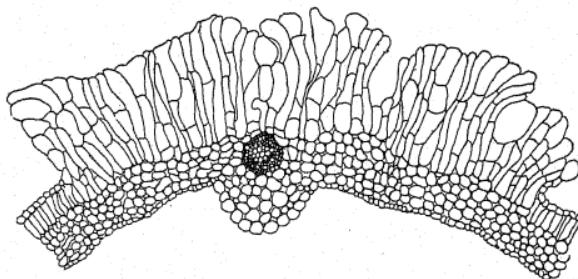


FIG. 9.—Large intumescence or patch formed by the union of several intumescences.

of cases, regards the lack of sufficient light to carry on rapid metabolism as an important factor in their formation. In "Die symptomische Bedeutung der Intumescenzen" (12), in which a number of cases

are described, he finds in every case, except one which developed under forced conditions in the greenhouse, that the intumescences were formed in the fall or winter, when the plants were in their rest period or approaching it. On account of the weak illumination during this time, the plants could not assimilate new building material and thus answered the stimulus of an over-abundant absorption of water by the formation of intumescences.

ATKINSON (1) explains the tomato oedema in a similar way. He says: "With the comparatively small amount of light carbon assimilation is lessened, so that the plant, under the forced conditions of growth, when it needs large quantities of carbohydrates, has to do with a really less quantity than is supplied in the open, when conditions for rapid growth are not so favorable and those for assimilation are improved."

KÜSTER (5) produced artificial intumescences on poplar leaves both in the dark and in the light, but found that when formed in intense light they occurred on the surface of the leaf which lay next the water. He suggested that in this case they did not form on the upper side on account of a more rapid transpiration, due to the strong light.

PRILLIEUX (8) observing the disease on pinks and NOACK (7) on grapes found that intumescences were produced only in weak light.

On the other hand, DALE (3) found that intumescences were not easily produced on *Hibiscus vitifolius* in poor light; were not formed under blue or green glass; but develop readily under red, yellow, or whitewashed glass and in bright light.

VIALA and PACOTTET (21) also strongly emphasize the fact that intumescences are only found in quantity on grape leaves directly under the glass, during periods of most brilliant illumination. These they regard as an effort of the plant to protect the leaf from "chlorovaporization and excessive transpiration" by the formation of a false palisade layer.

According to VON SCHRENK (9), intumescences are formed in the Missouri Botanical Garden only under these last-named conditions.

It is a matter of considerable interest, therefore, to determine the conditions under which intumescences are formed in the potato plant.

On March 12, 1906, thirty-six pots of potatoes were planted in

the greenhouse, to be used for the purpose of experiment. Eighteen of the potatoes were planted whole, while in the other pots were placed pieces containing two or three eyes. All of the potatoes were of good size and healthy. By April 4 several shoots had come up some distance from the ground and were covered with the bell jars.

#### EXPERIMENT I

Four pots of young plants with a few rather small young leaves were placed under bell jars in a well-lighted position in the greenhouse. The temperature of the air within the jars at the time of the setting-up of the experiment was 88° F. For three or four days after it was damp and cold, with only intermittent periods of sunshine. The temperature within the jars averaged about 65° F. In five to six days, after the temperature had increased to 68° F., intumescences began to appear.

Pot 1. Three shoots (from a cut potato), 205, 172, 128<sup>mm</sup> high respectively; all had rather small leaves.

Pot 2. Two shoots (from a cut potato), both 62<sup>mm</sup> high, with young leaves.

Pot 3. One shoot (from a whole potato), 36<sup>mm</sup> high, with young leaves.

Pot 4. Two shoots (from a whole potato), 32 and 61<sup>mm</sup> high, with small leaves.

#### Results

No. 1. After six days intumescences began to appear on all three plants and to assume the characteristic appearance. They were scattered over most of the leaves of the plant and were very numerous on two or three of them. Four days later they had become dry and blackened, but in two days more new ones had appeared.

No. 2. In five days intumescences had appeared on both shoots, although they were rather few and scattered over various leaves.

No. 3. In five days intumescences had formed, but not profusely.

No. 4. In five days a few intumescences had formed.

Intumescences were thus formed upon all of the shoots, but were more numerous on the shoots of pot 1. This was probably due to the fact that these plants were farther developed than the others and consequently did not grow so fast. The other plants were very young and grew rapidly, and in consequence made use of more water in growth than the other shoots. A discussion of the connection between growth and the formation of intumescences will be taken up later in this paper.

## EXPERIMENT II

At the same time as the preceding experiment, four plants were placed in darkness. The other conditions of temperature and moisture were the same. The bell jars were covered with black cloth to exclude the light.

Pot 5. One shoot (from a cut potato), 133<sup>mm</sup> high, with small leaves.

Pot 6. Two shoots (from a cut potato), 80 and 100<sup>mm</sup> high, with small leaves.

Pot 7. Three shoots (from a whole potato), 84, 63, and 25<sup>mm</sup> high, with small leaves.

Pot 8. Two shoots (from a whole potato), 63 and 25<sup>mm</sup> high, with small leaves.

*Results*

No. 5. In darkness eleven days and no intumescences formed.

No. 6. In darkness eleven days and no intumescences.

No. 7. In darkness eleven days; in six days two or three intumescences were found on one leaf.

No. 8. In darkness eleven days and no intumescences.

These plants were all small and their stems grew very rapidly in the dark. In the three cases where intumescences were not produced, it was thought that this might be due to the rapidity of growth. Accordingly a more developed plant was chosen and placed under the same conditions.

Pot 9. Two shoots (from a whole potato), 105 and 120<sup>mm</sup> high, with large, well-developed leaves. In two days a few intumescences had formed upon one of the shoots.

It will thus be noted that on two of the plants placed in darkness a few intumescences were formed. It was suggested that this might possibly be due to the fact that the bell jars were not absolutely light tight. Accordingly another pot was placed under the same conditions and the bell jar covered with several thicknesses of cloth to prevent light from filtering through the folds.

Pot 10. Two shoots (from a whole potato), 180 and 96<sup>mm</sup> high, with large leaves. No intumescences were formed.

These plants, after having been taken from darkness, were left in the light and allowed to develop their leaves. They were then covered with bell jars in the light and after eight days all the shoots, excepting those of pot 10, had developed intumescences.

## EXPERIMENT III

The next question which arose was in regard to the effect of increasing the absorption of water by the roots. Pots were placed over the warm water pipes, keeping the soil at a temperature of 70° F. The position of the plant was under whitewashed glass in the greenhouse.

Pot 11. One shoot (from a whole potato), 60<sup>mm</sup> high, with many large leaves.  
Pot 12. Two shoots (from a whole potato), 65 and 60<sup>mm</sup> high, with many large leaves.

*Results*

No. 11. After four days many intumescences were produced on many leaves.  
No. 12. After four days intumescences formed on nearly all the leaves and were very numerous on some of them. These intumescences continued to appear for several days.

## EXPERIMENT IV

Two plants were kept under the same conditions as in the preceding experiment, with the exception of being darkened by covering their bell jars with black cloth.

Pot 13. Two shoots (from a whole potato), 102 and 62<sup>mm</sup> high, with small leaves.  
Pot 14. Two shoots (from a whole potato), 185 and 130<sup>mm</sup> high, with large leaves.  
No intumescences were formed after ten days on either plant; the growth, however, in both cases was very rapid.

## EXPERIMENT V

The absorption of water was lessened by keeping the roots cold. Ice was kept packed around the pots, making an average temperature of the soil about 58° F. The pots were cut off from the air above in the bell jar by a thick blotting-paper covering. The air in the bell jars was kept warm by the sun (about 78° F.) during the days while the experiment was going on. A saturated atmosphere was produced within the jars by the evaporation from dishes of water.

*In light*

Pot 15. Two shoots (from a cut potato), 120 and 105<sup>mm</sup> high, with large leaves.  
Pot 16. One shoot (from a cut potato), 190<sup>mm</sup> high, with large leaves.

*In darkness*

Pot 17. One shoot (from a cut potato), 130<sup>mm</sup> high, with large leaves.

Pot 18. Two shoots (from a cut potato), 105 and 150<sup>mm</sup> high, with large leaves.

Although the plants were kept under these conditions for ten days, no intumescences were formed. The leaves on the plants in the dark became yellow and the plants finally died.

## EXPERIMENT VI

Four pots of plants were placed in a shaded portion of the greenhouse over warm water pipes, where their roots would be kept warm.

Pot 19. Two shoots (from a whole potato), 177 and 187<sup>mm</sup> high, with large leaves.

Pot 20. Two shoots (from a whole potato), 150 and 170<sup>mm</sup> high, with large leaves.

Pot 21. One shoot (from a cut potato), 85<sup>mm</sup> high, with rather small leaves.

Pot 22. One shoot (from a cut potato), 105<sup>mm</sup> high, with rather small leaves.

*Results*

In four days the leaves of nos. 19 and 20 were nearly covered with intumescences. Pots 21 and 22 also produced them, but they were not as numerous as on the preceding, and the growth of the plants was more rapid.

From this set of experiments it appears that intumescences appear in light and in the shade when the plants are abundantly supplied with water and their transpiration is reduced. They are found still more abundantly when their roots are kept warmer. Intumescences are not formed in complete darkness nor in light or darkness when the absorption of the roots is lessened by cold. Intumescences were not produced in any case on plants which were not covered by the bell jars.

At the end of this series of experiments all the plants used, as well as those grown outside the bell jars, were cut down and allowed to grow up again. Healthy shoots came up from all the plants excepting those which had been kept in darkness and had been killed by the rotting of their leaves and stems. In about ten days a few of the plants had reached a sufficient height to cover with the bell jars and the preceding experiments were repeated.

## EXPERIMENT VII

Plants placed in sunlight and covered May 7.

Pot 23. Two shoots (from a cut potato), 140 and 120<sup>mm</sup> high, with large leaves

Pot 24. Three shoots (from a whole potato), 150, 170, 180<sup>mm</sup> high, with large leaves.

In five days both plants showed many intumescences scattered over the leaves. No. 23 was particularly badly affected. There was a cold spell May 7-12, with temperature ranging from 62°-68° F., and a cloudy sky most of the time. On May 12 it became bright and warm, with a temperature of 72° F. Intumescences were then formed in great numbers.

#### EXPERIMENT VIII

Two plants were placed in darkness at the same time, with the other conditions similar to those of the preceding experiment.

Pot 25. Two shoots (from a cut potato), 180<sup>mm</sup> with large leaves, and 70<sup>mm</sup> with small leaves.

Pot 26. Three shoots (from a whole potato), 240, 170, 170<sup>mm</sup>, with large leaves.

In five days no intumescences were formed and the leaves became yellow and decayed.

#### EXPERIMENT IX

Two plants were placed under whitewashed glass, with their roots kept warm.

Pot 27. Two shoots (from a cut potato), 150 and 150<sup>mm</sup> high, with large leaves.

Pot 28. Two shoots (from a cut potato), 200 and 220<sup>mm</sup> high, with large leaves.

These were covered with bell jars on May 12, a bright warm day with temperature 72° F. In two days intumescences had formed very numerously on both plants. Plant no. 27, as shown in the photograph (*fig. 1*), was very heavily covered with intumescences, both on leaves and stem.

#### EXPERIMENT X

Two plants were placed in the dark, with their roots kept warm.

Pot 29. Two shoots (from a whole potato), 170 and 220<sup>mm</sup> high, with large leaves.

Pot 30. Two shoots (from a cut potato), 160 and 200<sup>mm</sup> high, with large leaves.

No intumescences were formed from May 12 to 15, but the leaves became yellow and decayed.

#### EXPERIMENT XI

Two plants were kept with their roots cold by packing the pots in ice.

Pot 31. In light; two shoots (from a whole potato), 200 and 190<sup>mm</sup> high, with large leaves.

Pot 32. In darkness; three shoots (from a whole potato), 150, 150, 150<sup>mm</sup> high, with large leaves.

These plants were left under these conditions from May 15 to 24, and no intumescences were formed. The temperature of the air in the bell jars was about 73° F., and of the soil 55° F.

#### EXPERIMENT XII

Four plants were placed in the shade, with their roots kept cold. The temperature of the roots averaged 56° F., and of the air above 73° F.

Pot 33. One shoot (from a cut potato), 135<sup>mm</sup> high, with rather large leaves.

Pot 34. One shoot (from a cut potato), 100<sup>mm</sup> high, with large leaves.

Pot 35. Three shoots (from a whole potato), 135, 100, 115<sup>mm</sup> high, with large leaves.

Pot 36. Two shoots (from a cut potato), 210 and 160<sup>mm</sup> high, with large leaves.

These plants were allowed to remain eleven days under these conditions and no intumescences were formed. Some of the leaves became yellow.

#### EXPERIMENT XIII

Four plants were placed in the shade, with their roots kept warm from the warm water pipes. The temperature of the air in the bell jars averaged about 84° F.

Pot 37. One shoot (from a whole potato), 170<sup>mm</sup> high, with large leaves.

Pot 38. Two shoots (from a cut potato), 120 and 100<sup>mm</sup> high, with large leaves.

Pot 39. Two shoots (from a cut potato), both 130<sup>mm</sup> high, with large leaves.

Pot 40. Three shoots (from a cut potato), 175, 150, 100<sup>mm</sup> high, with large leaves.

In four to five days intumescences had formed in very great numbers over all four of these plants.

On May 25, shoots which had come up a third time from the same lot of potatoes were covered with bell jars.

#### EXPERIMENT XIV

Two pots were placed in sunlight.

Pot 41. One shoot, 150<sup>mm</sup> high, with large leaves.

Pot 42. One shoot, 200<sup>mm</sup> high, with a slender stalk and small leaves.

In seven days no intumescences were formed.

## EXPERIMENT XV

Two pots were placed in the dark.

Pot 43. Two shoots, 200 and 230<sup>mm</sup> high, with large leaves.

Pot 44. Two shoots, 300 and 300<sup>mm</sup> high, with small leaves.

In seven days no intumescences were formed.

## EXPERIMENT XVI

Two plants were placed in the shade.

Pot 45. Three shoots, 200, 180, 160<sup>mm</sup> high, with small leaves.

Pot 46. Two shoots, 200 and 220<sup>mm</sup> high, with small leaves and a slender stalk.

In seven days no intumescences were formed.

## EXPERIMENT XVII

Two plants were placed under whitewashed glass and their roots kept warm.

Pot 47. Two shoots, 240 and 100<sup>mm</sup> high, with small leaves, but having flower buds.

Pot 48. Three shoots, 60, 150, and 200<sup>mm</sup> high, with small stalks and leaves.

In seven days no intumescences had formed.

That the intumescences did not form at all in this last series of experiments affords a suggestion as to their cause in the first set of young plants. The high osmotic pressure in the cells of the leaf is very probably brought about by the glucose in the leaves, which is furnished in great abundance to the young growing parts from the starch in the tuber, and is also assimilated in light by the leaves. In the case of these last experiments, the shoots which had been sent up the third time from the same tubers would thus be poorly supplied with glucose and the osmotic tension in the cells would not be so great. SORAUER (20), in a recent paper on the formation of internal intumescences on *Cereus nycticalis*, found a great abundance of glucose in the affected tissue and considered this to be the osmotically active substance which produced abnormal enlargement of the cells when a great amount of moisture was supplied. In the tomato ATKINSON (1) suggested that this substance is some organic acid, the production of which is increased by the low temperature and lessened light conditions under which the oedema developed. It is therefore very probable that the intumescences in the young potato

shoots are due to the presence of glucose when the other conditions of a humid atmosphere and an abundance of water for absorption are present.

As before stated, intumescences were not formed in complete darkness, but were developed in very great numbers in the shade and in the light, when supplied with an abundance of water and the root conditions were favorable for absorption. They were not formed on the young growing tips or on the old leaves which had ceased to grow. The explanation would therefore seem to be connected with the phenomenon of growth. It is probable that they did not form in total darkness because here the plants were stimulated to a rapid elongation of the cells in the stem and could thus take care of the abundant water supplied. At the same time, the glucose would not be so abundant as in plants growing in the light, since no assimilation takes place in darkness. For the same reason they did not form on the growing tips of the plants. They were formed in the sunlight and in the shade because elongation of the cells is not so rapid as in the dark and carbon assimilation is active. The plants could not thus take care of the water supplied fast enough, and the abundance of glucose in the growing leaves brought about the abnormal turgescence which caused the intumescences. That the intumescences were not found on the old mature leaves was probably due to the fact that these leaves had stopped growing and the cell walls were firmer, while less glucose was supplied to the cells from the tubers. When the tubers were healthy, intumescences formed equally well on plants from whole and cut potatoes. It would seem, therefore, that bright light is not necessary to the formation of intumescences in the case of the potato plant, nor does it act as a stimulus to their formation. This stimulus comes from the increased absorption of water and lessened transpiration with an abundance of glucose, and when enough light is present to produce the conditions for normal growth and assimilative activity.

To obtain a more accurate knowledge of the light intensity, in the various places in the greenhouse where the plants were placed, Wynn's photographic exposure meter was used. This instrument is manufactured for the use of photographers in measuring light intensity. The light values are obtained by exposing sensitized paper for a

time sufficient to bring it to the shade of a painted standard. The time which is necessary to produce the given shade may then be compared with the time taken to make the standard, which in this case was two to three seconds in bright sunlight at noon. In these experiments four seconds were necessary to produce the standard shade directly under the glass in sunlight, fifteen seconds under the whitewashed glass, and forty seconds in the shade. If the light value of the standard is represented by 1, the proportional light values under the three conditions given would be as follows:

Sunlight	Shade	Whitewashed glass
1.33	13.33	5.00

This method of obtaining light values is described by CLEMENTS (2, pp. 49-64). In order to test the power of new shoots from fresh potatoes to form intumescences, a new set of thirty-six potatoes was planted June 5; and June 25 the shoots were covered by the bell jars.

#### EXPERIMENT XVIII

In sunlight with an average temperature of  $79^{\circ}$  F.

Pot 48. Three shoots (from a whole potato), 240, 150, 150<sup>mm</sup> high, with large leaves.

Pot 49. Four shoots (from a cut potato), 320, 340, 300, 280<sup>mm</sup> high, with large leaves.

Pot 50. Three shoots (from a whole potato), with large leaves, 280, 230, 230<sup>mm</sup> high.

Pot 51. Two shoots (from a whole potato), 200 and 240<sup>mm</sup> high, with large leaves.

Pot 52. Four shoots (from a whole potato), 200, 240, 200, 220<sup>mm</sup> high, with large leaves.

After two days intumescences had begun to form, and in three days they had formed on all the plants excepting no. 52. Although this was left covered for a few days longer, intumescences did not form.

#### EXPERIMENT XIX

Five plants were placed in the shade, with an average temperature of  $73^{\circ}$  F.

Pot 53. Four shoots (from a cut potato), 240, 280, 210, 260<sup>mm</sup> high, with large leaves.

Pot 54. Three shoots (from a whole potato), 260, 230, 270<sup>mm</sup> high, with large leaves.

Pot 55. Three shoots (from a whole potato), 120, 130, 150<sup>mm</sup> high, with large leaves.

Pot 56. Four shoots (from a whole potato), 250, 240, 220, 100<sup>mm</sup> high, with large leaves.

Pot 57. Four shoots (from a whole potato), 130, 160, 170, 190<sup>mm</sup> high, with large leaves.

In two days intumescences had begun to form on all the plants. In three days they were very numerous over all the plants.

#### EXPERIMENT XX

Five plants were placed in darkness, with an average temperature of 79° F.

Pot 58. Four shoots (from a cut potato), 300, 250, 250, 270<sup>mm</sup> high, with large leaves.

Pot 59. Three shoots (from a cut potato), 280, 300, 290<sup>mm</sup> high, with large leaves.

Pot 60. Three shoots (from a whole potato), 300, 250, 140<sup>mm</sup> high, with large leaves.

Pot 61. Three shoots (from a cut potato), 230, 130, 150<sup>mm</sup> high, with large leaves.

Pot 62. Three shoots (from a cut potato), 240, 220, 200<sup>mm</sup> high, with large leaves.

In five days none of the plants had developed intumescences, but they were very badly decayed from the excessive warmth and moisture.

#### EXPERIMENT XXI

At the same time some of the old plants, the second and third shoots from potatoes planted previously, were placed under bell jars. They all had rather small leaves and slender stems and were from 200 to 300<sup>mm</sup> high. Three were placed in the shade and three in the sunlight and all failed to produce intumescences.

#### EXPERIMENT XXII

A few shoots of potatoes were stimulated with the various copper salts used by VON SCHRENK (9) in producing this disease on the cauliflower, to see if they would have a like effect upon the potato plant. The leaves of two shoots were stimulated with the following salts: copper ammonium carbonate, copper sulfate, copper acetate, copper nitrate, copper chlorid.

At the same time two shoots of the old plants were stimulated with each of these salts. All of these plants were left uncovered. After twenty-four hours burnt patches appeared on all the shoots wherever the sprays struck. The entire tissue through the leaf to

the under side in these patches was killed. No intumescences were formed on the old plants. On the new plants in two days a few intumescences formed as follows. On the first shoot sprayed with copper nitrate a few intumescences were produced over the veins. On the second shoot a few were formed on the surface. On one shoot sprayed with copper ammonium carbonate a few intumescences appeared at the tip of a leaf. The potato plants which were used, however, were not very healthy ones and developed from poor, shriveled, small tubers. These shoots would not form intumescences under bell jars when supplied with water, which fact was probably due to the leaves being poorly supplied with glucose. Had healthy young plants been used, it is very possible that intumescences would have been readily produced.

#### SUMMARY

Intumescences are produced on young potato plants from good tubers, when transpiration is checked and the roots are absorbing water. Their formation is increased by conditions which favor an increased absorption of water, as a warm soil, and is prevented by the opposite condition of a cold soil.

Intumescences are formed abundantly both in bright light and in weak light, which shows that light acts as a stimulus to their production only so far as it provides for the normal metabolism of the shoots and leaves.

Intumescences are not produced in total darkness, as this condition favors the rapid elongation of cells in the stem, which can thus make use of an increased supply of water. At the same time there is less of the osmotically active substance present in the leaves in darkness as photosynthesis is not taking place.

The abnormal state of turgescence of the hypertrophied cells is probably due to the osmotic action of glucose, assimilated in part by the leaf, but principally supplied to it from the underground tuber, so richly provided with starch.

In conclusion, I wish to acknowledge the many helpful suggestions of Professor ATKINSON, under whose direction this work was undertaken.

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GRAVITY AS A FORM-STIMULUS IN FUNGI  
CONTRIBUTIONS FROM THE HULL BOTANICAL LABORATORY  
XCII

HEINRICH HASSELBRING

(WITH THREE FIGURES)

The directive influence of gravity in determining the position of the hymenium in the higher Basidiomycetes is a matter of common observation. The fruit-bodies of these fungi are universally oriented so that the hymenium is exposed toward the earth's surface. If the normal position of the fruit-body is changed, either by accident or by experimentation, readjustment takes place by which the hymenial surface is again brought into a horizontal plane, provided of course that such readjustment is not hindered by other factors. Interesting cases illustrating these phenomena are given by ATKINSON<sup>1</sup> and also by WATERS.<sup>2</sup>

The methods by which the horizontal orientation of the pileus is brought about are two. First, in all stalked forms, such as the Agaricaceae, and some of the Polyporaceae and Hydnaceae, the stalks are negatively geotropic, and by their curvature always bring the pileus into a horizontal position. This property was first observed by HOFMEISTER<sup>3</sup> in some of the Agaricaceae. The phenomena may be observed in all stalked forms growing on the sides of trees or stumps. Here the stems curve upward until the pileus is brought into the normal position. Second, in forms which have no stalk, and this applies especially to sessile forms of Polyporaceae, the orientation of the hymenium is brought about by the growth of the trama-plates themselves, for, as SACHS<sup>4</sup> has shown, the trama-plates of the hymeno-

<sup>1</sup> ATKINSON, G. F., Mushrooms, edible, poisonous, etc. pp. 13-17. 1900.

\_\_\_\_\_, Some wood-destroying Fungi. Geol. Surv. of Louisiana, Rep. 1899: 333-338. pls. 7.

<sup>2</sup> WATERS, C. E., Geotropism of *Polyporus*. Plant World 7:224. 1904.

<sup>3</sup> HOFMEISTER, W., Ueber die durch die Schwerkraft bestimmten Richtungen von Pflanzenteilen. Jahrb. Wiss. Bot. 3:92. 1863.

<sup>4</sup> SACHS J., Experimental Physiologie. p. 93. 1865.

phore, not only in the Polyporaceae but also in the Hydnaceae and Agaricaceae, are positively geotropic. Since in non-stalked forms adjustment of the whole pileus is obviously impossible, the downward growth of the hymenophore alone is responsible for the orientation of the hymenium. If the position of the growing pileus is changed, readjustment to the new condition is accomplished solely by the change in direction of growth of the elements of the trama-plates and the growing, undifferentiated elements of the pileus. This often results in the formation of new pilei growing at various angles from the old pileus (see ATKINSON, *l. c.*).

In such types as the Polyporaceae it is evident that the form of the fungus depends to a large extent on the growth of the trama-plates making up the hymenophore. Since the mode of growth of these elements is greatly influenced by gravity, the question naturally arises as to what extent the form which these fungi assume is determined by the responses of the elements to the influence of gravity. To determine the effect of gravity as a form-stimulus, klinostat experiments were conducted with a number of fungi from different families. As a rule sticks and branches upon which sporophores were growing were brought into the laboratory for experimentation. Although a considerable number of plants were thus tried, only a few gave results, owing, no doubt, to the fact that the conditions of growth of these forms are not sufficiently well understood so that they can be cultivated with certainty. From a few forms interesting results were nevertheless obtained. The experiments are here first described, and this is followed by a discussion of the significance of the results obtained.

In October 1905 a small fruit-body of *Polystictus cinnabarinus* growing on a stick was placed in a moist chamber and rotated on the horizontal axis of the klinostat until the middle of December. The fruit-body was a little over 1<sup>cm</sup> in diameter, with few pores, and growing at the margin. During the experiment its growth was slow, although the plant appeared to remain healthy. At the end of the experiment the plant was about 2<sup>cm</sup> in diameter. The new growth, however, was not confined to the margin as in stationary fruit-bodies, but owing, no doubt, to the still embryonic condition of the fruit-body growth was resumed over the entire surface, resulting

in a pulvinate fruit-body. The most remarkable result of the experiment, however, is this: the fruit-body was no longer differentiated into a sterile dorsal and fertile ventral surface; the whole pulvinate body was covered with tubes characteristic of the hymenial surface of this fungus. At the present time it is scarcely possible to distinguish the original dorsal and ventral surfaces of the specimen, which has been preserved in alcohol. With the suspension of the differentiation of dorsal and ventral surfaces, all tendency of the fruit-body to produce the normal shelf-like form is of course lost.

On October 9, 1906, a stick with several growing fruit-bodies of *P. cinnabarinus* was placed in a moist chamber as before, and revolved in a dark chamber until December 15. At the same time several other sticks with growing fruit-bodies were similarly arranged and revolved, without excluding the light. Of all the fruit-bodies used in this experiment only one rather large specimen in the dark chamber showed any uniform growth.<sup>5</sup> This one had grown on the upper side of a dead branch, so that it had a somewhat rotate form, attached at the center. The sporophore was about 3.5<sup>cm</sup> in diameter. The growth of this plant on the klinostat was extremely slow, so that it did not enlarge much. New growth occurred along a zone on the margin, extending back for about 1<sup>cm</sup> over the dorsal surface. The result was a tumid thickening of the otherwise somewhat acute margin of the pileus. Here again the whole growing zone showed the incepts of numerous pores characteristic of the hymenial surface. That these were not more developed, as in the former case, is due to the unfavorable conditions of growth, for it should be remembered that the other fruit-bodies used in this experiment, whether growing in light or darkness, made scarcely any growth at all, although they remained fresh and normal to all appearances.

Among the Agaricaceae experiments were carried out with *Schizophyllum commune* and some species of Coprinus.

Pieces of a maple branch containing mycelium of *Schizophyllum* were placed on the klinostat as in former experiments. At the beginning of the experiment any traces of sporophores visible on the

<sup>5</sup> At the end of the experiment it was found that several of the smaller fruit-bodies had developed well-marked pores in patches, where growth had been resumed on the margin and on the upper surface.

branch were removed. It was therefore certain that if any fruit-bodies appeared, they had developed from their earliest stages on the klinostat. The precaution is important, since the fruit-bodies of the Agaricaceae are completely differentiated during their earliest stages of growth. Beyond these stages the parts, therefore, can no longer be subject to formative stimuli. On several of these sticks the fruit-bodies developed within about a month. They appear as small outgrowths resembling simple forms of *Clavaria*, and attain a length of about 1<sup>cm</sup>. Early in their development a cup-like depression appears at the summit, and within this the rudimentary lamellae are formed, radiating from the center. The fruit-bodies at this stage have the form of a short trumpet about 1<sup>cm</sup> long. The outside has the characteristic appearance of the dorsal surface of the normal fruit-body. By subsequent growth the margin expands uniformly at first, but later unequally, so that the resulting fruit-body is a lobed disk, 2-3<sup>cm</sup> in diameter, and stalked at the back. The curious fact brought out by this experiment is that the lamellae develop on the inner side of the trumpet- or funnel-shaped fruit-body. The pileus, therefore, appears to be attached at the back, with the lamellae developing on the morphologically upper side.

To study the effect of gravity on some more specialized form, dung-cultures were set up for *Cóprinus*. These were continually rotated so that the entire development of the fungi from the germination of the spore to the maturation of the fruit-body took place on the klinostat. The primordia of the fruit-bodies were thus at no time subject to the normal influence of gravity. Several crops of *Cóprinus* developed on the cultures, but the fruit-bodies were normal in every way. Their direction of growth was perpendicular to the substratum, the actual direction being horizontal, since the pots were rotated on the horizontal axis of the klinostat.

The experiments with the foregoing three types show the existence of a marked difference in the degree to which the form of these types is modified by or depends upon the stimulus of gravity. Of the plants studied, *Polystictus* is the more plastic, and is consequently more influenced by gravity. Here the differentiation of the fruit-body into the sterile dorsal and fertile ventral surface depends on the direct or indirect action of gravity. In *Schizophyllum* the tissue

differentiation is independent of the external stimulus of gravity, but the general form of the plant is somewhat modified in a way to be considered later. In the highly differentiated *Coprinus* the morphogenic influence of gravity is not evident.

The sporophores of all the higher fungi are made up of a complex mass of hyphae whose growth is so correlated that the final result is the definitely organized fruit-body. These hyphae must be considered to a large extent as independent individuals, for they have organic connection only in so far as they arise from common branches many cell generations back in the tissue, or in so far as they are united by subsequent fusion which is comparatively rare. The elements, nevertheless, grow in unison to form the sporophore. As the organization of the sporophore is in the main the result of successive changes in the direction of growth of elements,<sup>6</sup> it seemed possible that this mode of growth might be determined by external stimuli, coupled of course with a changing capacity of the elements for reacting to those stimuli. For example, if gravity alone were considered, the hyphae organizing the pileus of mushrooms might be plagiotropic while forming the cap, those on the lower side becoming positively geotropic to organize the lamellae, while the hyphae organizing the hymenium would again be plagiotropic. Of course it is not supposed, and the experiments bear this out, that any case would yield to so simple an analysis as this, for it is still possible that the plant has within itself an inherited tendency toward a definite organization, and even if this is not admitted the common chemotropic and thigmotrophic actions of the hyphae upon each other play an important part. That direct external stimuli also play an important part is shown by the non-development of *Lentinus lepideus*<sup>7</sup> in the dark. The caps of some species of *Coprinus*<sup>8</sup> also are dependent upon light

<sup>6</sup> ATKINSON, G. F., The development of *Agaricus campestris*. BOT. GAZETTE 42:241-264. pls. 6. 1906; the older literature is reviewed here.

ALLEN, C. L. The development of some species of *Hypholoma*. Ann. Myc. 4: 387-394. pls. 3. 1906.

<sup>7</sup> REINKE, J., Ueber Deformation von Pflanzen durch äussere Einflüsse. Bot. Zeit. 62:81-112. pl. 1. 1904; see the literature given here.

BULLER, A. H. R., The reactions of the fruit-bodies of *Lentinus lepideus* Fr. to external stimuli. Annals of Botany 19:427-438. pl. 4. 1905.

<sup>8</sup> BREFELD, O., Untersuchungen über Schimmel-pilze 3:87-97, 114-115. 1877.

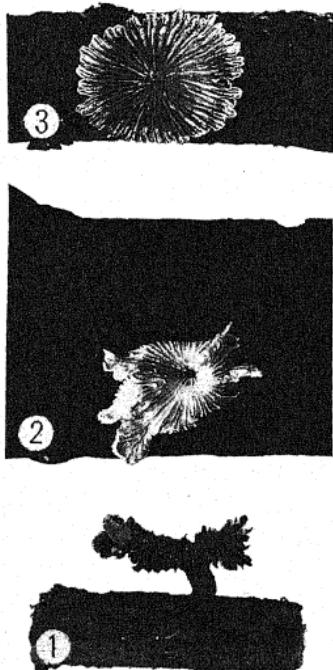
for their development. In these cases the development of the pileus is retarded or suppressed, so that it is probable that here light acts as something more than a tropic stimulus. BULLER (*l. c.*) has recently shown, however, that the lamellae of *Lentinus* develop first and most vigorously on the lower side of the young fruit-body when it arises obliquely from the substratum, and that this unilateral development is due to the effect of gravity. Here we have a change in form due to the effect of gravity. It is probable that the eccentricity of the stalks of tree- and stump-inhabiting forms is directly due to such action of gravity, for when some of these forms, like *Pleurotus ulmarius*, develop on the upper side of a fallen trunk they are symmetrical.

In *Polystictus cinnabarinus*, as in other shelf-forms, the fruit-bodies increase in size by growth at the margin. The hyphae near the lower side of the growing margin turn downward and produce the hymenophore. The epinastic growth (if this term, originally applied only to dorsiventral organs, may be employed) of these hyphae is due to their positive geotropism, for on the klinostat they grow uniformly in all directions. The organization of the hymenophore itself is not dependent upon gravity, for when this force is equalized all the hyphae form hymenial tissue. The differentiation of the fruit-body into an upper sterile and a lower fertile region is also due ultimately to the influence of gravity. All the hyphae are potentially fertile, but the development of a hymenium by the upper hyphae is inhibited finally by gravity. How this sterilization is brought about is another question. It is probable that the direct cause is to be sought in the mechanical or chemotropic influences of the hyphae upon each other. Since experiments were conducted both in light and in dark, it is evident that light is not a factor in this case.

The form of the more highly organized *Schizophyllum* is influenced in a less degree. It is evident that the general organization of the fungus is quite independent of the stimulus of gravity. There is one interesting phase, however, that needs to be considered. The fruit-bodies of this fungus are typically dorsiventral and laterally attached, yet on the klinostat they originate as small funnel-shaped bodies with the lamellae on the inside of the funnel; and when allowed to develop fully, the funnel expands and develops into a lobed disk,

stalked at the back and bearing the gills on the upper surface (*figs. 1 and 2*). It will be seen that the gills are not, as in all other Agaricaceae, on that surface of the pileus which is continuous with the stipe, *i. e.*, the lower surface, but on the opposite side, which corresponds to the dorsal surface of other mushrooms. In this respect *Schizophyllum* stands alone, at present, among the Agaricaceae. The growth of the fungus on the klinostat enables us to interpret the structure of the normal dorsiventral fruit-body.

The youngest primordia arise in the form of minute cups. If these are allowed to develop on the klinostat, the tendency of the pileus to develop equally on all portions shows itself. If, however, the fungus grows out more or less horizontally from the substratum, as it normally grows, the lower half of the pileus ceases to develop, while the upper half grows into a fan-shaped fruit-body. The pileus may be regarded as a funnel with the gills on the inside, having its lower half cut away and the upper half flattened out so as to expose the gills in their normal position toward the earth. The inhibition of growth of the lower half is ultimately due to gravity, since on the klinostat all portions develop equally. The same effect is produced when the fruit-bodies grow on the lower side of sticks, as they often do in nature. Such specimens expand symmetrically on the margin, producing a rotate fruit-body attached at the back



*Schizophyllum commune*. FIG. 1.—Stalked symmetrical fruit-body grown on the klinostat. FIG. 2.—Lateral view. FIG. 3.—Fruit-body developed on lower side of branch lying on the ground, showing symmetrical structure. X 1.

and with the lamellae radiating in all directions from the center (*fig. 3*).

In the highly differentiated *Coprinus* no formative influence of gravity could be observed. It is evident that here the organization of the fruit-body depends largely upon internal causes or upon stimuli not yet sufficiently analyzed.

In conclusion, it follows from the foregoing observations, on three widely separated forms of the Basidiomycetes, that although gravity has no apparent effect on the organization of the hymenophore,<sup>9</sup> it has a marked influence on determining the configuration of the fruit-body of some forms. This effect is most marked in the more primitive forms, which are thereby shown to be the more plastic. In the more highly differentiated forms this effect of gravity disappears. The effect of gravity on the configuration of *Polystictus* is due partly to the positive geotropic property of the hymenophore, and partly to the fact that the differentiation of the fruit-body into sterile and fertile tissue depends on the action of gravity. When this force is removed, the fruit-body assumes the resupinate or a pulvinate form characteristic of the simplest types of the Basidiomycetes.

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<sup>9</sup> This term applies only to the parts immediately bearing the hymenium, *i. e.*, the gills, spines, etc., and not, as it is sometimes used, to the whole fruit-body.

## ASCOGENOUS FORMS OF GLOEOSPORIUM AND COLLETOTRICHUM

C. L. SHEAR AND ANNA K. WOOD

An accurate knowledge of the life-histories of fungi is of the greatest importance, not only to taxonomy, but especially to plant pathology. As many parasitic fungi possess two or more fruiting stages, in some cases occurring on different hosts, it is necessary to have a full knowledge of their complete course of development in order to know how they are reproduced and distributed, and to devise adequate methods of combating or preventing them.

The fungi producing the diseases known as anthracnoses, which attack a great variety of wild and cultivated plants, have been described chiefly as species of *Gloeosporium* and *Colletotrichum*. The positive connection of these conidial forms with ascogenous stages was, so far as we are aware, first demonstrated by means of pure cultures by Professor G. F. ATKINSON, as reported by Miss STONEMAN<sup>1</sup> in 1898. FÜCKEL<sup>2</sup> in 1869 gave *Gloeosporium juglandis* Lib. (*Marsonia juglandis* (Lib.) Sacc.) as the spermagonial form of *Gnomonia leptostyla* (Fr.) Ces. & De Not., which occurs on *Juglans regia*. It has uniseptate conidia, and the ascospores are also uniseptate, which facts seem to indicate that it is not congeneric with the organisms with which we are concerned at present. FÜCKEL'S statement in regard to the connection between these two forms was based simply upon the association of the two on the same leaf, and was merely a guess. KLEBAHN<sup>3</sup>, however, has recently demonstrated by pure cultures and infection experiments that FÜCKEL was correct in this case. Other forms studied by KLEBAHN, which have passed under the name of *Gloeosporium*, are *G. nervisequum* (Fück.)

<sup>1</sup> STONEMAN, B., The development of some anthracnoses. BOT. GAZETTE 26: 69. 1898.

<sup>2</sup> FÜCKEL, L., Symbolae Mycologicae 123. 1869.

<sup>3</sup> KLEBAHN, H., Zusammenhänge von Ascomyceten mit Fungis imperfectis. Cent. f. Bakt. 15<sup>a</sup>:336. 1905.

Sacc.<sup>4</sup> and *G. Ribis* (Lib.) Mon. & Desm.<sup>5</sup> The first of these he connects with *Gnomonia veneta* (Sacc. & Speg.) Kleb., which has uniseptate ascospores, and hence is not congeneric with the anthracnoses, whose ascospores are not septate. In the case of *G. Ribis*, KLEBAHN<sup>6</sup> claims to have demonstrated by infection experiments and by pure cultures that its ascogenous stage is a discomycete, which he has described as *Pseudopeziza Ribis*. It is very remarkable if conidial forms so similar as to be placed by taxonomists in the same genus should have ascogenous stages belonging to such different, distinct groups as the Pyrenomycetes and the Discomycetes. Miss STONEMAN (*l.c.*) in 1898, CLINTON<sup>6</sup> in 1902, VON SCHRENK and SPAULDING<sup>7</sup> in 1903, SHELDON<sup>8</sup> in 1905 and 1906, and SCOTT<sup>9</sup> in 1906 have worked with organisms from various hosts which have usually passed under the name of *Gloeosporium* or *Colletotrichum*, and which are undoubtedly congeneric with the plants treated of in this paper.

We may say, in the beginning, that neither of the names just mentioned is tenable for any of the plants to be considered here, as these names were originally applied to species belonging to other genera than those to which they are applied by SACCARDO and other recent mycologists. The name *Glomerella*, of SPAULDING and VON SCHRENK, will be used here, as it is the only one which we are absolutely certain at present belongs to this group of organisms. There is little doubt, however, that there are older valid generic names which have been applied to either the conidial or ascogenous stages of these

<sup>4</sup> KLEBAHN, H., Untersuchungen über einige Fungi imperfecti und die zugehörigen Ascomycetenformen. II. Jahrb. Wiss. Bot. 41:515. 1905.

—, Untersuchungen über einige Fungi imperfecti und die zugehörigen Ascomycetenformen. III. Zeits. Pflanzenkrank. 16:65. 1906.

<sup>6</sup> CLINTON, G. P., Apple rots in Illinois. Agr. Exp. Sta. Bull. no. 69. 1902.

<sup>7</sup> SCHRENK, H. VON, and SPAULDING, P., The bitter-rot of the apple. U. S. Dept. Agr., Bur. Pl. Ind., Bull. no. 44. 1903.

<sup>8</sup> SHELDON, J. L., Concerning the identity of the fungi causing an anthracnose of the sweet-pea and the bitter-rot of the apple. Science N. S. 22:51. 1905.

—, The ascigerous stage of *Gloeosporium Psidii*. Science N. S. 21: 143. 1905.

—, The ripe rot or mummy disease of guavas. W. Va. Agr. Ex. Sta. Bull. no. 104. 1906.

<sup>9</sup> SCOTT, W. M., The control of the apple bitter-rot. U. S. Dept. Agr., Bur. Pl. Ind., Bull. no. 93. 1906.

fungi, but until we have had opportunity to study the types of the genera to which they may belong, we prefer to use the name *Glomerella* for at least all the species which are known to have ascogenous stages congeneric with *Glomerella rufomaculans* (Berk.) Spauld. & v. Schrenk.

The first published work on the life-history of these fungi was that of Miss STONEMAN, in 1898; she worked with twelve forms from as many different hosts, and succeeded in producing in pure cultures both the conidial and ascogenous stages in four cases, and a doubtful fifth, as follows:

*Glomerella cingulata* (Stoneman) Spauld. & v. Schrenk, the conidial stage occurring on privet (*Ligustrum vulgare*); *G. piperata* (Stoneman) Spauld. & v. Schrenk, the conidial stage found on pepper, *Capsicum* sp.; *G. cincta* (Stoneman) Spauld. & v. Schrenk, the conidial stage occurring on an orchid, *Maxillaria picta*, and provisionally referred to *Colletotrichum*; *G. rubicola* (Stoneman) Spauld. & v. Schrenk, the conidia from red raspberry, *Rubus strigosus*; *G. (?) Vanillae* (Stoneman) Spauld. & v. Schrenk, the conidia from the cultivated vanilla.

Miss STONEMAN referred all these species to a new genus, which she named *Gnomoniopsis*. This name was untenable, however, having been previously used by BERLESE<sup>10</sup> for another genus of fungi. CLINTON (*l. c.*) in 1902, seems to have been the first to obtain in pure cultures the ascogenous stage of the Gloeosporium causing anthracnose or bitter-rot of the apple. VON SCHRENK and SPAULDING (*l. c.*) in 1903, and SCOTT (*l. c.*) in 1906, also worked with the apple Gloeosporium and made additions to our knowledge of its life-history and conditions of development. SHELDON (*l. c.*) in 1906, reported the successful growth in pure cultures of the ascogenous stage of the guava anthracnose or ripe-rot, *Glomerella Psidii* (Del.) Sheldon. These six forms are the only ones whose life-histories have heretofore been determined by pure culture methods, so far as we have been able to learn from published records.

The present writers have studied a number of forms and have succeeded in growing both the conidial and ascogenous stages from eight different hosts, as follows:

*Gloeosporium rufomaculans* (Berk.) v. Thümén, from the cultivated grape, *Vitis* sp.; *G. fructigenum* Berk., from the apple; an apparently unnamed Gloe-

<sup>10</sup> BERLESE, A. N., *Icones Fungorum* 1:93. 1892.

osprium from the cranberry, *Vaccinium macrocarpum*; *G. elatiae* Cooke & Massee, from the leaves of the rubber plant, *Ficus elastica*; a form from the locust, *Gleditschia triacanthus*, which does not appear to have been reported before; one from *Ginkgo biloba*, also not heretofore reported; *Colletotrichum gossypii* Southw., from cotton; and *C. Lindemuthianum* (Sacc. & Magnus) Bri. & Cav., from the cultivated bean.

Of these eight only the one from the apple has had its ascogenous form reported heretofore. Miss STONEMAN worked with the anthracnose of the bean, but was unable to produce the ascogenous stage.

All the forms mentioned have been grown by the writers in pure cultures, and the ascogenous stages produced, in most cases, several times. The original cultures were usually obtained by the transfer to culture media of conidia or portions of the tissue of the host containing the hyphae of the fungus. We have been unable as yet to tell definitely what the factors are which control the production of the ascogenous form. Repeated experiments with various culture media, under various conditions of light, temperature, and moisture, seem to indicate that these are not factors of great importance. Once having obtained a race, strain, or generation which produces ascogenous perithecia in cultures, it can be successfully grown on various media and under various conditions for several generations.

The medium which we have found best adapted for use and which has produced the most abundant and luxuriant growths of ascogenous forms is sterilized corn meal. We have found by repeated experiments that if a culture from any particular acervulus or group of acervuli does not produce an ascogenous stage on corn meal at 75° to 85° F., it is useless to experiment further with material from the same source. Fresh material from other specimens must be tried until a race, strain, or generation is found which will produce its ascogenous form. It has been suggested that the nearness or remoteness of origin of the conidia from ascospores may be an important factor in determining the production of an ascogenous stage. The few experiments we have made along this line are not conclusive, and more data must be obtained before results can be reported.

Heretofore forms occurring on different hosts have been generally regarded as distinct species. The study which we have made of the eight forms just mentioned leads to the conclusion that they cannot

be successfully segregated as species upon the basis of morphological characters. Though we have examined a great deal of material, of both conidial and ascogenous stages, from various pure cultures, as well as from the natural hosts, we are unable to find any differences of sufficient constancy or importance to make it possible to distinguish species or even varieties except the host be considered. Miss STONE-MAN describes and figures most of her species as having perithecia with a distinct neck or beak. We have very rarely seen in any of our cultures as conspicuous a beak as she figures. In all other characters, however, the forms we have studied agree with hers. The conidia of these fungi are well known to be quite variable in size and shape, and it is frequently possible to find in a single acervulus conidia varying in length from 10 to 25  $\mu$  and in thickness from 3.5 to 6  $\mu$ , so that the range of variation in a single culture will usually cover the range of variation found in any of the forms we have studied. This is not only true of the conidia, but is also true of the ascogenous forms. The greatest variation found in the size of ascospores is from 9 to 24  $\mu$  by 4.5 to 8  $\mu$ , the usual measurements being about 14 to 18  $\mu$  by 5 to 6  $\mu$ . In shape the asci vary somewhat, being usually sub-cylindrical or clavate. The ascospores vary but little in shape and appearance of contents. They are usually slightly curved or allantoid, having the contents in fresh specimens regularly granular, with a more or less distinct hyaline zone at the center, and when fully matured are of a light greenish-yellow color. This color, however, is hardly noticeable except in mature specimens. From this it will be seen that these organisms cannot be satisfactorily distinguished by their morphological characters, so far as the conidia and asci are concerned. Some of the conidial forms have been separated heretofore upon the basis of the presence or absence of dark setae in the acervulus. This, as has been pointed out by other writers, is also an uncertain and variable character. We have found setae in the acervuli in one part of a pure culture, whereas in other parts of the same culture they were absent. We have also found this same variation in acervuli occurring upon leaves, especially in the form found upon the cranberry.

The question remains whether these organisms can or should be separated as physiological species. The answer depends upon

whether the forms are restricted to their individual hosts, or can be successfully transferred from one host to another. The work already done by HALSTED,<sup>11</sup> SOUTHWORTH,<sup>12</sup> and others in transferring various forms occurring upon different fruits from one host to another by inserting the conidia in the tissue of the host, or by applying them to the surface of mature fruits does not seem to us to be conclusive. A mature fruit, especially when the surface is ruptured, might be regarded simply as a culture medium, and the fact that these organisms will grow upon it does not prove that they would grow upon the plant as a parasite. The only conclusive test of this matter must be made by careful infection experiments, using fresh, living, and growing plants controlled by checks. SHELDON<sup>13</sup> reports having done this in the case of the anthracnose of the apple, having succeeded in infecting the leaves of the sweet-pea by transferring conidia from the apple. These experiments, in connection with others which have been reported to us verbally, seem to indicate that some of these forms are not physiologically distinct. In some cases at least the evidence indicates that plants which appear free from disease are already infected, the fungus apparently being in a dormant or more or less inactive condition and awaiting favorable conditions for development. In the anthracnose of cotton and bean, the disease is known to be transmitted by diseased seed. It is desirable, therefore, that plants used for infection experiments should be grown from healthy disinfected seed under such conditions as to prevent the possibility of infection from other sources. In the present state of our knowledge, perhaps it may be best to regard the various forms we have studied as varieties of one species.

The bodies frequently found in nature and in cultures, which have been called by some chlamydospores and by others appressoria, show no specific characters. While their function, perhaps, may be primarily that of appressoria or hold-fast organs, as maintained by HASSELBRING<sup>14</sup> and others, they are often produced under conditions

<sup>11</sup> HALSTED, B. D., Laboratory study of fruit decays. Report N. J. Agr. Exp. Sta. 1892: 326. 1893.

<sup>12</sup> SOUTHWORTH, E. A., Ripe rot of grapes and apples. *Jour. Myc.* 6:164. 1891.

<sup>13</sup> SHELDON, J. L., Concerning the identity of the fungi causing an anthracnose of the sweet-pea and the bitter-rot of the apple. *Science N. S.* 22:51. 1905.

<sup>14</sup> HASSELBRING, H., Appressoria of the anthracnoses. *BOT. GAZETTE* 42:135. 1906.

which seem to indicate that they may have some other function as well. They have frequently been found in our cultures, and occur also in cranberries that have been destroyed by the cranberry anthracnose. They vary considerably in size and shape, and the germ pore which is attributed to them is frequently indistinct or wanting.

Most writers who have studied the ascogenous stages of these fungi have described them as without paraphyses. SHELDON<sup>15</sup> has recently mentioned finding paraphyses in ascogenous perithecia produced from the apple. We have occasionally found organs surrounding the outer portion of the mass of asci which bear a close resemblance to the paraphyses of certain other pyrenomycetous fungi. A careful study of these organs shows that they are not intermingled with the asci, but are about the outer portion of the mass, next to the wall of the perithecium. In many instances they suggest aborted or malformed asci. In any case they would be more correctly called periphyses than paraphyses. Their rare occurrence would seem to indicate that they are of no great importance for taxonomic purposes.

Since these organisms have been found to produce ascogenous perithecia in cultures, it would be expected that they would also produce them under natural conditions upon their host plants. The conidial forms are so numerous and so widely distributed that if the ascogenous forms occur often they would probably have been found and described by mycologists before now. DELACROIX<sup>16</sup> describes *Glomerella (?) Artocarpi* as found on *Artocarpus* leaves associated with *Gloeosporium* and *Colletotrichum*. The description and figures agree well with our ascogenous forms. Since beginning this investigation we have examined carefully various plants attacked by anthracnose in the hope of finding ascogenous perithecia under natural conditions. Thus far, however, we have been able to find them in only two cases; that of the apple, which had already been reported by CLINTON (*J. C.*), and that of the rubber plant, *Ficus elastica*. The ascogenous perithecia are frequently found in abundance upon fallen leaves of the rubber plant, which have been attacked by the anthracnose, *Gloeosporium elasticae* Cke. & Mass. They agree in

<sup>15</sup> SHELDON, J. L., The ripe rot or mummy disease of guavas. W. Va., Agr. Exp. Sta., Bull. 104: 310. 1906.

<sup>16</sup> DELACROIX, G., Bull. Soc. Myc. France 21:198. 1905.

every respect with those which are produced in pure cultures, except that the perithecia in the pure cultures are more or less overgrown with the vegetative hyphae, and this we find to be generally true in the case of cultures of perithecia of other fungi, which under natural conditions are borne within the tissues of the host. There are several genera of pyrenomycetous fungi which have characters so nearly like those of *Glomerella* that they cannot be very satisfactorily distinguished from it, and we are of the opinion that some of the species already referred to these genera are really the ascogenous stages of anthracnoses. Of such genera we may mention *Physalospora*, *Phomatospora*, and *Guignardia*. As already stated, for the present it seems advisable to treat these organisms as varieties of the oldest species, *Glomerella rufomaculans* (Berk.) Spauld. & v. Schrenk, which was originally described from specimens from the grape.

U. S. DEPARTMENT OF AGRICULTURE  
Washington, D. C.

## LICHEN DISTRIBUTION IN THE SANTA CRUZ PENINSULA, CALIFORNIA

ALBERT W. C. T. HERRE

The Santa Cruz peninsula, California, forms a rough triangle extending in a northwesterly direction from Monterey Bay on the south to the Golden Gate at the north, a distance of ninety miles or more. In breadth it varies from about thirty-five miles at the widest portion to perhaps six miles near the northern end. Laved on the west by the Pacific, its eastern boundary is formed by San Francisco Bay and the broad, originally treeless Santa Clara-San Benito valley; while at the southeast the Pajaro cuts its way through the range and separates it from the Gabilan range.

Rising from sea-level along most of its border, it reaches at its highest point an elevation of 3793 feet and embraces within its limits every variation between the dense, unbroken redwood forest and the bare, sea-girt rock, the cloud-swept mountain crag, and the vast expanse of monotonous salt marsh.

Separated thus from the adjacent country and having such a diversified surface, it forms a natural biological region, well known both to the zoologist and the botanist. Though lichens are notorious for their wide geographical distribution, a study of the Santa Cruz lichens shows that they too are affected by the comparative isolation of the peninsula.

In addition to its possession of a number of endemic forms, one of the most interesting features of the lichen flora of the Santa Cruz peninsula is the commingling of boreal or alpine, temperate, and tropical species. At the same time, there is a remarkable absence of certain lichens which we should naturally expect to find in the region. That no species of the genus *Graphis* should be found, and that *Cladonia rangiferina* and all forms of the genus *Stereocaulon* are absent, is an anomaly that as yet seems baffling, since forms ordinarily associated with all these are common.

But the mingling here of lichens typical of different phytogeographical regions can be explained, I believe, by a study of the

climatology of the Pacific Coast and of the Santa Cruz peninsula in particular, recognizing at the same time that the area under discussion is really an aggregate of a number of minor biological areas, each possessing distinct physiographic, faunal, and floral features, and marked by its own typical lichen species.

Most people, in a general way, have the idea that California has a subtropical climate, and that name is often applied to it. In reality, owing to its great area and wonderfully diversified topography, California has many climates, but the typical Californian climate is, more than that of any other part of the United States, a true cool temperate climate; and it is to the rather cool but remarkably equable temperature conditions that we primarily owe the interesting intermixture of lichens typical of unlike life-regions.

As typical of the temperature of the Santa Cruz peninsula I quote the following averages for the twelve months of the year, as fixed by observations at San Francisco extending over more than half a century: "January, 52.2° F.; February, 52°; March, 54°; April, 55°; May, 57°; June, July, and August, each 59°; September, 60.8°; October, 60°; November, 56°; December, 52°."

These temperatures will favor the boreal or alpine forms such as *Gyrophora polyphylla*, *Evernia vulpina*, *Lecidea caeruleo-nigricans*, *Rhizocarpon geographicum*, and others of like character; while at the same time they will permit of the migration from warmer regions of various species of *Ramalina*, *Dendrographa*, *Dirina*, *Lecanactis*, *Schismatomma*, etc.

While the temperature conditions of the whole Santa Cruz peninsula are quite uniform, the annual rainfall shows great variation in different localities, ranging from 50 or 60 inches at Boulder Creek, and even more in the Big Basin, to 13 or 14 inches at Stanford University.

For detailed statistics on this whole matter, however, one must refer to Professor ALEXANDER MCADIE's valuable work upon *The Climatology of California*.

A study of the lichen flora of the Santa Cruz peninsula shows that it is more or less sharply divided into a number of biologic areas, which may be roughly classified as the maritime, foothill, chaparral, mountain-forest, and mountain-peak areas.

The maritime area is sharply defined, more so in fact than any of the others, and includes not only a narrow strip of land all along the coast but also all of the northern tip of the peninsula down to and including the San Bruno mountains. Most of this area is of course of low elevation, but in the northern part of San Mateo County, where the broken hills of San Francisco culminate in San Bruno mountain, it extends to an altitude of more than 1300 feet.

This maritime region possesses the most equable climate of the peninsula, and also perhaps the highest daily average of humidity. It is characterized by relatively cold, windy, and foggy summers, the fogs mitigating the dryness of the rainless summer months; the winters are comparatively clear, sunshiny, with less precipitation than the redwood forests farther inland, and much warmer than in the other areas. At several points along the coast a whole winter often passes without the temperature once falling to 32° F.

This area may (in a large way) be considered as a part of the belt which extends southward to the tropics, and beginning again on the coast of Peru reaches far to the south of Valparaiso, Chile. It is a region in the main of slight rainfall and very moderate range of temperature, and it is therefore not surprising that we find a number of rock or earth lichens common alike to the shores of California, Peru, and Chile, yet unknown except on the Pacific coast of the Americas.

I have no means of knowing how far to the north of the Golden Gate these forms go, but most of them, including those originally described by TUCKERMAN from the San Francisco Bay region, seem to become more luxuriant as we go farther south, though I have seen none from below Guadalupe Island, Lower California. This would seem to indicate that such forms may be regarded as migrants from regions lying nearer to the tropics, and that the Golden Gate is very near to their northern limit, if indeed they go beyond it. In this category we may place most of the characteristic lichens of the maritime region.

The characteristic lichens of this area do not occur in any of the other areas, and in that respect it is the most strongly marked of them all. Some of the more important are as follows:

*Arthopyrenia halodytes* (Nyl.) Oliv.; *Buellia halonia* (Ach.) Tuck.; *Dendrographa minor* (Tuck.) Darbish.

*Dirina franciscana* A. Zahlbrückner, n. sp.—Thallus effuse, of thick, rounded, irregular tuberculate areoles, uniform crustaceous, sub-cartilaginous, forming heaped patches. Color varying from yellowish or brownish yellow to a dingy ashy gray, the last most common; K—; C—.

Apothecia numerous, of medium size, rounded elevated, sessile; surface of disk minutely granular, ashy-gray pruinose; the thalline margin thick, prominent, white, obtuse, soon flexuous, often intricately so; epithecium dark,  $56 \mu$  high; hypothecium black, thick,  $140 \mu$  high, blue or bluish with I; paraphyses typical of the genus, thecium wine-red with I; ascii clavate, straight or curved, long-stalked,  $\frac{70-134.5}{16.8-22.4} \mu$ ; spores eight, colorless, fusiform, straight or slightly curv.d, quadrilocular,  $\frac{23.8-33.5}{5-8} \mu$ .

On rocks 50 to 75 feet above the sea at Point Lobos, San Francisco, growing with *Dendrographa minor*.

Near *Dirina repanda* of Europe and Northern Africa, but with a thicker and differently colored thallus and with different spores.

*Lecanactis Zahlbrückneri* Herre, n. sp.—Thallus effuse, of small, irregular (sometimes plicate) squamules which at first are scattered but soon become a thick, uniform, tartaceous crust. Color a more or less evident rose-pink which soon fades out in herbarium specimens, leaving them whitish or ashy gray.

Apothecia small, round, sessile, becoming convex; black, the disk gray-pruinose, but eventually naked; the proper margin prominent but finally excluded. Epithecum dark or black, thick,  $45$  to  $50 \mu$  high, blue with I; hypothecium black, broad,  $42$  to  $60 \mu$  high; paraphyses typical of the genus, thecium brick- or vinous-red with I; ascii clavate, straight or curved, sometimes pointed at tip,  $\frac{78-106}{16.8} \mu$ ; spores eight, colorless, fusiform, straight or slightly curved, quadrilocular,  $\frac{19.6-28}{5-7} \mu$ .

Rare on maritime rocks, 50 to 75 feet above the sea, at Point Lobos, San Francisco. Associated with *Dendrographa minor*, *Arthopyrenia halodytes*, and *Trenteophelia* sp.

*Lecanora Bolanderi* Tuck.; *L. pinguis* Tuck.; *L. phryganitis* Tuck.; *Opegrapha saxicola* Ach.; *Parmelia Herrei* A. Zahlbr.; *P. Borri* Turn.; *P. perlata* (L.) Ach.; *Pertusaria multipuncta* (Turn.) Nyl.; *Physcia erinacea* (Ach.) Tuck.; *Placodium coralloides* Tuck.; *Ramalina ceruchis* (Ach.) DeNot.; *R. ceruchis* cephalota Tuck.; *R. combeoides* Nyl.; *R. homalea* Ach.; *Rinodina radiata* Tuck.

The foothill region comprises all that territory between San Francisco Bay and the mountains proper, south of San Bruno mountain, and extends from sea-level to a height of approximately 1000

feet. On the Pacific side it is either very narrow or non-existent, but on the eastern side of the peninsula it extends as far south as Monterey Bay. A park-like oak forest is characteristic of the lower part of the region, and on these trees is a lichen flora remarkable for its variety and luxuriance. While but few lichens are confined to this area, many reach here their greatest development. Probably every tourist is struck with the appearance of the "lace lichen," *Ramalina reticulata* (Noehd.) Krempelh., which decks the trees much as "Spanish moss" clothes the live oaks of the Gulf Coast. Other noticeable lichens are:

*Collema nigrescens* (Huds.) Wainio; *C. vespertilio* (Ltf.) Wainio; *Evernia prunastri* (L.) Ach.; *Lecanora subfuscata* (L.) Ach.; *L. varia* (Ehrh.) Nyl.; *Lecidea granosa* Tuck.; *L. granulosa phyllizans* A. Zahlbr.; *Leptogium chloromelum stellans* Tuck.; *L. palmatum* (Huds.) Mont.; *Pannaria lepidiota cyanolepra* Tuck.; *P. lepidiota coralliphora* Tuck.; *Parmelia soredica* Nyl.; *Placodium cerinum* (Hedw.) Naeg. & Hepp.; *P. ferrugineum* (Huds.) Hepp.; *Physcia pulverulenta isidiigera* A. Zahlbr.; *Ramalina Menziesii* Tuck.; *Theloschistus lychneus laciniosus* (Schaer.).

The chaparral belt is not sharply delimited by contour lines or elevations, but depends upon climatic as well as physiographic features, the characteristic angiosperms of the chaparral being well-marked xerophytes. Hence we may have chaparral occurring on dry, barren hill-tops as low as four hundred feet, while we find more or less of it all the way to the summit of the Santa Cruz mountains. Very few lichens are exclusively chaparral, but where the conditions were favorable to such movement many conspicuous species have migrated from the adjacent forests. Certain components of the chaparral, such as the manzanitas, are almost wholly devoid of lichens ordinarily, because of their thick evergreen foliage and exfoliating bark. Others, as *Ceanothus*, *Cercocarpus*, and the scrub oaks, are conspicuous for their dense growth of lichens. The chief lichens in this formation are:

*Cetraria californica* Tuck.; *Cladonia verticillata* Hoffm.; *Lecanora pallens* (L.) Schaefer.; *Parmelia enteromorpha* Ach.; *Ramalina farinacea* (L.) Ach.; *Usnea plicata* (Ach.) Nyl.; *U. dasypoga* (Ach.) Nyl.; *U. florida* (L.) Ach.

The typical mountain forest of the Santa Cruz peninsula is the redwood and Douglas spruce formation, though much of the forested

area is given over to a growth of oaks, madrone, and other trees. In the redwood formation, sheltered by the dense canopy overhead, the lichens are but little affected by the frosts of winter, while the heavy summer fogs counteract the effect of the summer drouths. Having such favorable life-conditions for lichens, we may regard this region as the distributing center from which the chaparral has been stocked, while the foothill area has also largely been supplied in the same way. Arboreal forms such as the Stictaceae and certain Parmelias reach their highest vegetative and reproductive development in the dense redwood forests; while many others living on the earth or mosses are fertile and well grown only in the forests at 2000 feet or above, though they may descend in the foothills to as low as 150 feet. In this connection it may be well to call attention to the fact that the forests cover a much smaller area than formerly. For the last half-century the ax of the lumberman and forest fires have been actively engaged in the process of deforestation, and another generation may see many of the characteristic plants of the redwood forest extinct except within the limits of the Big Basin, which the state has purchased and set aside for a state park. Some of the more striking lichens of the forests are:

*Alectoria jubata* (L.) Tuck.; *Cetraria ciliaris* (Ach.) Tuck.; *C. lacunosa stenophylla* Tuck.; *Cladonia macilenta* (Hoffm.) Nyl.; *Leptogium albociliatum* Desmaz.; *L. californicum platynum* Tuck.; *Nephromium helveticum* Ach.; *N. lusitanicum* (Schaer.) Nyl.; *N. tomentosum rameum* Nyl.; *Parmelia enteromorpha* Ach.; *P. perforata* (Wulf.) Ach.; *P. tiliacea* (Hoffm.) Ach.; *Peltigera canina membranacea* (Ach.) Nyl.; *Pertusaria Wulfenii* DC.; *Lobaria pulmonaria* (L.) Hoffm.; *L. scrobiculata* (Scop.) DC.; *Sticta anthraspis* Ach.; *S. fuliginosa* (Dicks.) S. Gray; *S. limbata* (Sm.) Ach.; *Sphaerophorus globosus* (Huds.); *Usnea longissima* Ach.; *U. californica* Herre.

By the mountain-peak area is meant the masses of exposed rock which may occur about the heads of heavily forested cañons, as well as the bare and rocky mountain peaks extending above the forests and chaparral. The lichens of this area are typically saxicolous, and while they may occur throughout the other formations wherever there is sufficient area of rock surface, some are found only along the highest mountains, swept by raw, bleak winds and exposed to a great diurnal as well as annual range of temperature. In summer this may rise to a sun temperature of 150° F. or more, and in winter may

descend to 20° F. or thereabouts, accompanied by more or less snow. These conditions of course can be met only by lichens specially adapted for such a life, of marked xerophytic structure, and there is no such enormous thalline development as is observed in the foothills or redwood formation. The lichen flora of this region, however, is not below that of any of the others in either number of species or of individuals. Important lichens of this region are:

*Collema plicatile* Ach.; *Gyrophora diabolica* A. Zahlbr.; *G. phaea* (Tuck.) Herre; *G. polyphylla* (L.) Turn. & Borr.; *Lecanora muralis* diffracta Fr.; *L. calcarea* (L.) Sommerf.; *L. sordida* (Pers.) Th. Fr.; *L. upsaliensis* Nyl.; *Lecidea caeruleo-nigricans* (Lightf.) Schaer.; *L. auriculata* Th. Fr.; *L. polycarpa* Fr.; *Rhizocarpon geographicum* DC.; *Parmelia conspurcata* (Schaer.) Wainio; *Verrucaria terebrata* (Mudd).

The lists published are merely some of the most conspicuous of each area and are by no means exhaustive. In the author's herbarium are more than 200 named species and sub-species of lichens which he has collected in the Santa Cruz region, with perhaps nearly a hundred more as yet unstudied or not satisfactorily determined. It is believed that the lichen flora of the region under discussion will be found to include more than 300 species and sub-species.

SAN JOSÉ, CALIFORNIA

## BRIEFER ARTICLES

### NEW NORMAL APPLIANCES FOR USE IN PLANT PHYSIOLOGY. IV<sup>1</sup>

(WITH TWO FIGURES)

In the three preceding articles I have described seven new pieces of normal apparatus devised for educational work in plant physiology; and below will be found accounts of two more, to be followed later by others. I call them normal appliances because they are designed and made expressly for their specific work, yield approximately accurate quantitative results, can be used with economy of time and effort, and are obtainable at any time from the stock of a supply company, which in the present instance is the Bausch and Lomb Optical Company of Rochester, N. Y. The development of this apparatus is part of the present movement toward the elevation of educational plant physiology to a higher plane of scientific logic, accuracy, and efficiency.

#### VIII. RESPIROMETER

Respiration is a universal, and the most important, process of organic nature, and hence demands effective demonstration in all biological courses. Its study is the more valuable in plant physiology because, while respiration is essentially identical in plants and animals, it can be investigated far more readily in plants. Its central and crucial fact, energy-release, cannot be directly demonstrated by any known method, but indirectly it can be proven and its amount determined through the identification and measurement of the gases absorbed and released in the process. For this purpose many arrangements have been described, of all grades from complex and precise to simple and inaccurate. The present new instrument is designed both to exhibit and to measure the gas exchanges in typical respiring material (*e. g.*, germinating seeds), and to accomplish this with considerable accuracy and convenience of manipulation (*fig. 1*).

The respirometer consists of three parts. First is the stoppered oval chamber for the seeds, with a water bulb at the bottom. Second is the measuring cylinder in open communication with the chamber, graduated from 75° to 100° of the combined capacity of itself and chamber, though the 75° mark is actually placed at 77° of the capacity. Third, and

<sup>1</sup> Continued from BOT. GAZETTE 41:213. March 1906.

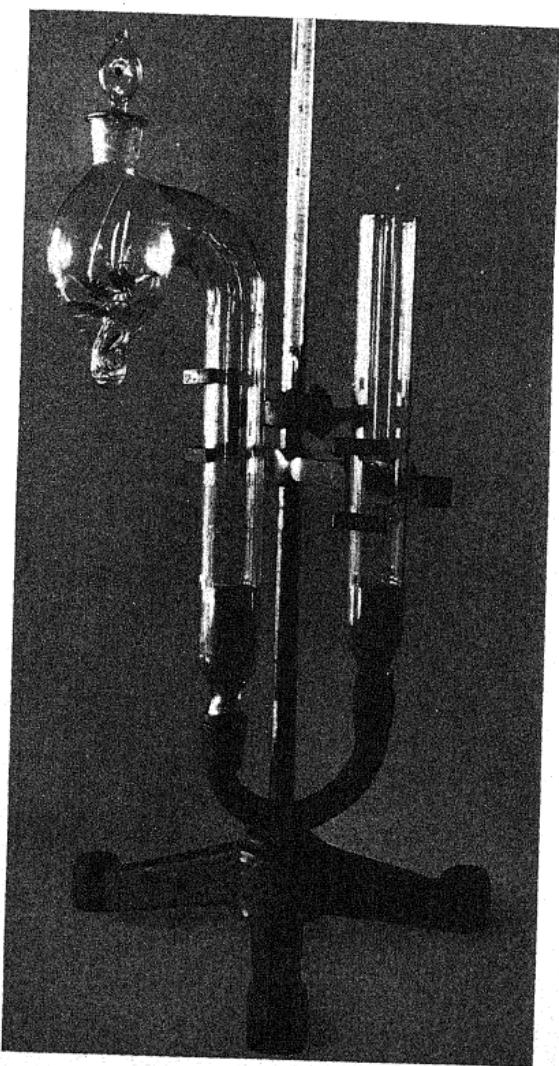


FIG. 1.—Respirometer.

communicating with the preceding through a slender rubber tube, is the reservoir cylinder, ungraduated but with index marks 25<sup>cm</sup> apart. Both tubes are supported vertically by any convenient laboratory clamps which permit the reservoir tube to be slipped up and down.

For demonstration purposes it is best to select seeds in which the oxygen absorbed and the carbon dioxid released are as nearly as possible equal in volume, *e. g.*, oats. Ten of these of average size are soaked, or better, are selected from a lot which have been started in a germinator until the roots are about 5<sup>mm</sup> long; they are then placed in the chamber, root ends down, just above the bulb, where they will stick if previously wetted. These occupy approximately 1<sup>cc</sup> of volume; and if now 1<sup>cc</sup> of water be placed in the bulb, there will be 2<sup>cc</sup> of these materials and 100<sup>cc</sup> of air, the composition of which is of course known, above the zero mark. Where greater accuracy is desired, it may be attained by first dropping the ten seeds into a proper measuring-glass, then filling this with water to the 2<sup>cc</sup> mark, and finally placing both seeds and water in the chamber. The index liquid to be used is now poured through the reservoir tube until it stands level at the 100<sup>cc</sup> mark of the graduated tube and at the upper index mark of the reservoir (or the lower when mercury is used). The stopper, properly lubricated, is inserted with its air opening matching that of the neck, and is then twisted, thus sealing the chamber without any compression of air. The apparatus is now shielded from light and placed under proper conditions for growth.

For the most effective educational demonstration it is desirable, though not necessary, to use two of the respirometers. In one is placed a strong solution of caustic potash, which rises steadily, as the seeds grow, to near the 21 per cent. mark and then stops. This proves that some gas has vanished from the tube, and the amount suggests oxygen. In the other mercury is used, and the constant level it maintains proves that for any gas absorbed by the plants an equal volume of some other has been released. Since however caustic postash absorbs only carbon dioxid, it is plain that the gas released must be carbon dioxid, while the gas absorbed is presumably oxygen. If it is thought this latter point still needs proof, it can be supplied by a third instrument using pyrogallate of potash. This promptly absorbs all oxygen, and the failure of the seeds to grow shows that oxygen is the gas absorbed in the other instruments. If but one instrument is available, it is best to use it with mercury, and, after the seeds have been growing for some three or four days, to make an analysis of the gas by the method earlier described<sup>2</sup> for the photosynthometer. The reservoir and

<sup>2</sup> BOT. GAZETTE 41:211. 1906.

rubber tubes are slipped off under water, allowing the mercury to run out, and are then used as a reagent tube, the reservoir being stoppered for the purpose.

When plant parts in which the respiratory ratio is not 0 are studied, a preliminary test is made to ascertain in which direction the ratio varies, and then the starting-level can be adjusted accordingly.

The usual corrections of course must be made. The gas pressure inside at the time of reading is equalized with the atmospheric pressure by sliding the reservoir tube up or down until the levels inside and out are equal. For very exact work it would be necessary to take account of the barometric pressures, but the slight error of this sort is negligible in demonstration. The temperature must either be made the same at the start of the experiment and the final reading; or else, as is readily possible, the change of volume due thereto must be calculated. Vapor tension should also be considered in exact work, but it is negligible in demonstration. After each use the instrument should be thoroughly washed clear of potash.

#### IX. NORMAL LIGHT SCREENS

In several phases of the study of photosynthesis it is necessary to apply some form of light or color screen to a leaf. To this end various arrangements are extemporized which, however inconvenient and time-consuming to prepare, are scientifically unexceptionable so long as they are physiologically accurate. But unfortunately some of these devices are logically fallacious and yield results which are only accidentally correct. This is true, for example, of that experiment common in elementary demonstration, where disks of cork, tin-foil, etc., are placed matching on opposite sides of the leaf in order to exclude light; in fact, they also exclude the equally indispensable carbon dioxide.

Screens correct in principle must allow for free access and exit of gases, but in their construction advantage may be taken of the fact that in ordinary leaves the stomata are either largely or wholly on the under surface, so that if this is left free the upper surface may be covered as closely as desired. It is upon this principle the two forms figured herewith (*fig. 2*) are constructed. The larger, designed to take an entire leaf of moderate size, consists of a wooden box readily adjustable for height and angle,  $5 \times 7 \times 1\frac{1}{2}$  inches (internal), white without and black within, separated lengthwise into two compartments with an intermediate space for petiole and midrib. The bottoms of the compartments are largely open, but so matched by diaphragms that air can enter freely but no direct light. Movable gratings of silk threads hold the leaf firmly but elastically against the glass cover,

which may be either two separate strips covering the compartments and therefore the halves of the leaf, or a single sheet  $5 \times 7$  inches in size. The cover may then carry tin-foil, cut with any desired pattern, gummed to its under surface; or it may carry vials of pure colors correlative to the light and dark spots of the chlorophyll spectrum (as was the case when the accompanying photograph was taken); or it may be replaced by a  $5 \times 7$  negative for Gardiner's striking starch-printing experiment. The arrangement does not of course permit as free access of carbon dioxide as the uncovered leaf enjoys, but this is only a matter of degree and does not affect the result when sufficient time is allowed for the experiment. In using

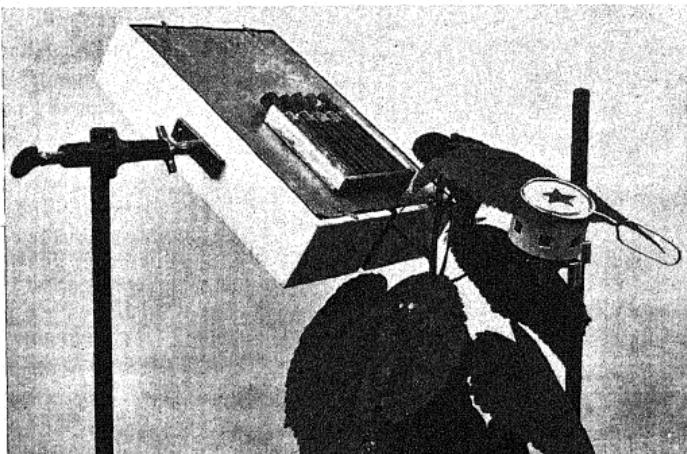


FIG. 2.—Normal light screens.

this, as any other, light screen it is desirable to expose the plants not to direct sunlight but to strong diffused light, which is nearer the optimum and has less heating effect. The leaves of course are subsequently blanched and treated with iodin in the usual manner.

The smaller screen, of a less range of usefulness but much cheaper, is made upon the same general principle except that it is constructed to clasp a portion of a leaf. A spring clip holds a glass disk against the upper surface of a leaf, which is supported below by a grating of threads stretched across the top of a ventilated dark box. The glass is removable from the clip and may carry a tin-foil screen cut with a pattern and gummed to its under surface as in the figure; or it may be used to hold a photographic

negative (film or glass) against the leaf, in either case permitting a very striking demonstration of the need for light in photosynthesis.

Both instruments appear to be correct in principle; both are convenient and efficient in manipulation; and they yield most satisfactory results.  
—W. F. GANONG, *Smith College, Northampton, Mass.*

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### IS THIS BIRCH NEW?

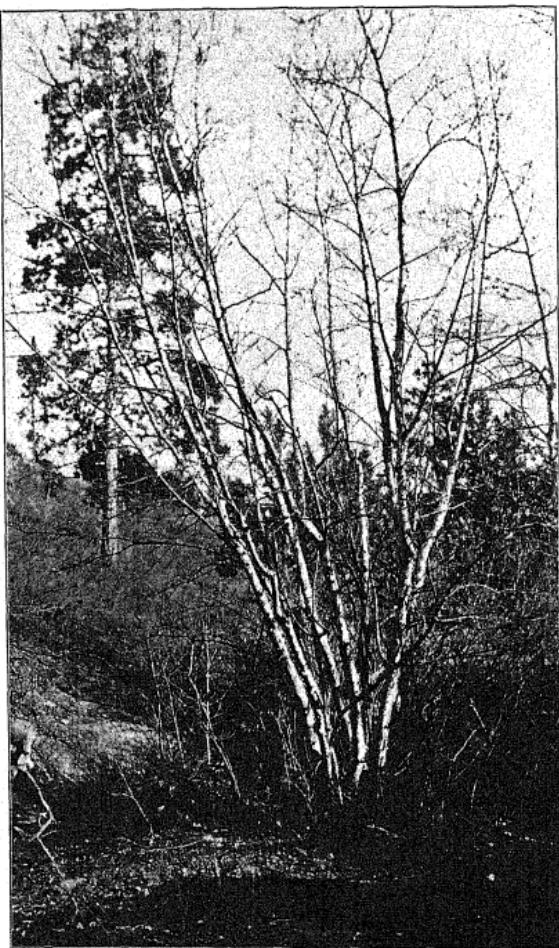
(WITH ONE FIGURE)

The paper birch (*Betula papyrifera* Marsh.) has never been reported from Colorado. The nearest region to Colorado from which authentic specimens have been obtained is probably the Black Hills in northwestern Wyoming and adjacent South Dakota. It was therefore a great surprise when Mr. D. M. ANDREWS of Boulder, Colorado, reported finding a birch which in many respects resembles *B. papyrifera*. He writes of it as follows: "About three miles from Boulder on the north slope of Green Mountain, altitude 6500-7000 feet, I find one hundred trees, more or less, of the species from which I send you specimens."

An examination of the material sent, which, by the way, was secured so late in the season that the specimens are not in good condition, indicates its affinity to *B. papyrifera*. It cannot be referred to the var. *cordifolia*, and it is even less like typical *B. papyrifera*. In the material so far available, however, it is difficult to point out specific distinctions. From the latter it differs in the darker bark, which on the trunk is marked by long transverse rifts (lenticels). The current season's twigs of ANDREWS' specimens are light brown, the two-year-old twigs more or less grayish, and the older branches reddish brown and distinctly marked by the white oval lenticels. The bark on the main trunks is silvery gray, materially darker than in *B. papyrifera*, though it peels readily into layers as in that species. The leaves do not seem to differ essentially, being either broadly cuneate or rounded at the base. The margin is rather sharply and irregularly serrate, though the teeth are not long. The apex is somewhat abruptly short-acuminate. The bracts of the fruit are deeply 3-lobed, the central lobe being longer and narrower than the lateral. The nut is oblong-obovate, with very thin wings nearly twice as broad as the body.

Its habit is perhaps its striking character. In *B. papyrifera* there is usually a single trunk, which branches above in true tree fashion. ANDREWS writes of his birch as follows: "You will notice the uniform tendency to form clumps or stools, which is true of quite young specimens. The

photograph of the older clump shows trunks about six inches in diameter, which seems to be the maximum, after which the old trunks die and are



*Betula*, n. sp. ?, showing characteristic habit; collected near Boulder, Colorado, by D. M. Andrews.

replaced by young sprouts from the same root. While the difference in size (between this and *B. papyrifera*) may be due to differences of environ-

ment, I do not believe it to be entirely so, and suspect that in cultivation they would remain distinct, age for age, *B. papyrifera* growing more rapidly."

Further studies another season may show other characters that will absolutely separate this newly found birch from *B. papyrifera*. Should such be the case, the writer will then propose the name *Betula Andrewsii* for this new find, in honor of Mr. ANDREWS, whose diligent studies and discriminating observations in the field are again recognized—studies the more valuable because the opinions formed in the field are usually verified in his experimental and nursery grounds at Boulder.—AVEN NELSON, *University of Wyoming, Laramie.*

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#### RHODODENDRON ALBIFLORUM WITH DOUBLE FLOWERS

While on a botanizing tour with Mr. J. G. JACK in British Columbia in the summer of 1904, we were staying several days at Glacier, the station of the Canadian Pacific R. R. close to the foot of the Great Glacier of the Selkirks, to explore the surrounding country. Just above the hotel in the Asulkan valley there is a grove of *Tsuga Mertensiana* and *T. heterophylla*, through which a path leads to the foot of the glacier. As I left this path to cross a thicket of small hemlocks to the bank of the Asulkan River, I noticed close to the brink of the river a large shrub of *Rhododendron albiflorum* in full bloom which at once attracted my attention, as all other shrubs of this species were past flowering; and I was very much astonished to find on coming nearer that it bore large white double flowers very much like those of the double cherries sometimes cultivated in gardens. It was a strange sight in these wild and rough mountain woods to see such blossoms which one associates involuntarily with the finished surroundings of a well-kept garden. What agency caused the origin of such a form? Close by grew the typical form, and there was nothing unusual in the place or position where the shrub grew, nor in the shrub itself aside from its double flowers. This is probably the first time that a double-flowered Rhododendron has been found in this country. Though reports of the occurrence of double-flowered plants in a wild state are not wholly lacking, they are nevertheless rare. In the European Alps *Rhododendron ferrugineum* has been found at least twice with double flowers, as reported by A. KERNER (*Oesterr. Bot. Zeits.* 15:285. 1865), who himself found in one locality a large number of shrubs with double flowers.

As in most double flowers, the cause of this teratological aberration is in this case petalody of stamens combined with a considerable increase

in the number of the staminal whorls. Calyx and corolla present the normal condition, while the stamens have taken the shape of petals. Though the ten stamens belong to two whorls, the petalodes are more or less connate at the base into 10-merous whorls; but the petalodes of all the whorls are in superposition, showing thus that they must be considered as consisting of alternate 5-merous whorls, and this is also apparent by the aestivation of the broader petalodes of the outer whorls; the first five petalodes, of course, are opposite to the corolla lobes, as the ericaceous flower is obdiplostemonous. The number of 10-merous whorls in each flower is about ten; the shape of the petalodes changes from the broadly oval of the outer ones to the oblong shape of the innermost petalodes. Even the carpels are changed into narrow oblong petalodes. Only in one case I found an incompletely developed anther adhering to one of the petalodes.

As it would be interesting to have this shrub in cultivation, though the horticultural value of this form is lessened by the rather difficult cultivation of the species, we sent rooted suckers to the Arnold Arboretum, but the plants did not survive, and concerning the fate of a few sent to a German nursery I know nothing. The old plant, however, is in all probability still there, and we may yet hope to see this handsome form introduced into cultivation. Dried specimens of it are preserved in the herbarium of the Arnold Arboretum.—ALFRED REHDER, *Arnold Arboretum*.

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#### PUCCINIA UPON MELOTHRIA

(WITH ONE FIGURE)

This rust was found in the neighborhood of West Raleigh, N. C., in the autumn of 1907 upon *Melothria pendula*. The host is not a common plant in this locality, only two plants being seen during an entire collecting season; one of these was thoroughly infected, the other not at all.

The species is of special interest on account of the comparative paucity of rusts affecting the cucurbits, as well as owing to the economic importance of this family. On the whole family as represented by the North American genera, either native or exotic, listed in SMALL'S *Flora of the South Eastern United States*, GRAY'S *Manual*, GRAY'S *Field, Forest, and Garden Botany*, and BRITTON and BROWN'S *Illustrated Flora*, there are mentioned in FARLOW'S *Host Index* no rusts at all. In SACCARDO there are six species, as follows: (1) *Uromyces Melothriæ* on *M. tomentosa* in Abyssinia; (2) *U. Cayaponiae* on *C. racemosa* in Africa; (3) *U. Hellarianus* on *Cayaponia* in Porto Rico; (4) *Puccinia Cucumeris* on *C. ficifolia* in Abyssinia; (5) *P.*

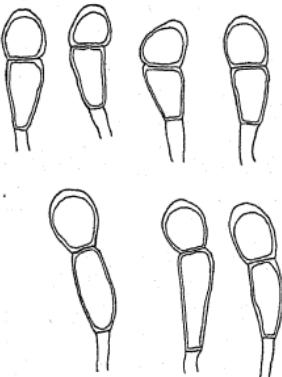
*Momordicae* on *M. cordijolia* in Natal; (6) *Aecidium Momordicae* on *Momordica* in Brazil.

The *Puccinia* upon *Melothria pendula* is different in shape, color, and size of spores from *P. Cucumeris* and from *P. Momordicae*, and seems clearly to be an undescribed species.

The germination of the teleuto-spores occurs freely in the older sori, placing the rust among the Leptopucciniæ. The following description is proposed.

*Puccinia Melothriæ*, n. sp.—III.  
Teleutosori: hypophyllous, crowded in irregular concentric circles, pustular, soon naked, pulverulent, cinnamon-colored (no. 32 Sacc.), cinereous on germination, ruptured epidermis inconspicuous; spots pale, indefinite. Teleutospores: hyaline to straw-colored (no. 26 Sacc.),  $40-52 \times 10-16 \mu$ ; slightly or not at all thickened at apex, often not constricted at septum, wall about  $2 \mu$  thick, smooth. Terminal cell usually round, rarely oblong,  $10-16 \times 12-20 \mu$ . Basal cell wedge-shaped,  $10-15 \times 20-30 \mu$ . Pedicil  $20-45 \mu$  long, hyaline. Germination immediate; that of the terminal cell by an apical promycelium, that of the basal cell by a lateral promycelium.

On *Melothria pendula* L., West Raleigh, N. C., 9-15-06, F. L. Stevens and J. G. Hall, type no. 471.—F. L. STEVENS, West Raleigh, N. C.



*Puccinia Melothriæ*, n. sp.

#### AN INSTANCE OF MUTATION

*Euphorbia marginata* Pursh, commonly called in Colorado "snow-on-the-mountain," is easily recognized by its white-margined upper leaves or bracts, and the five white appendages of the floral glands. In the splitting up of the old genus *Euphorbia*, it has been referred to a genus *Dichrophyllum* Kl. & Garcke, and distinguished from *Tithymalus* Adans by the characters just mentioned. In *Tithymalus* there are but four glands, with rudiments of the fifth. All authors seem to agree that *E. marginata* has five glands; "three" in BRITTON and BROWN, *Ill. Flora* 2:376, is a mere misprint, as the figure shows. Having recently had occasion to examine *E. marginata* at Boulder, Colorado, I have been astonished to find that four glands is normal in this locality, and a plant with five glands

has not been seen. One large plant deviated still more, showing mostly three glands, and the white margins of the leaves very narrow. I wrote to Professor AVEN NELSON about this, and he replies "all Colorado specimens do not show the reduction in number which you mention, and some of the specimens are quite devoid of white on the margins of many of the leaves." We seem to have a clear case in which a new race has been formed by mutation, or more precisely the *meristic variation* of BATESON. Incidentally it seems fairly evident that the genus *Dichrophyllum* cannot be maintained; so we have:

*Tithymalus marginatus* (*Euphorbia marginata* Pursh, 1814);

*T. marginatus tetramerus*, var. nov., the Boulder race with four glands.—  
T. D. A. COCKERELL, *University of Colorado, Boulder.*

## CURRENT LITERATURE

### MINOR NOTICES

**Das Pflanzenreich.**<sup>1</sup>—Part 27 contains the Polemoniaceae by A. BRAND, a family of peculiar interest to North American botanists. The author defines two subfamilies, COBAEOIDEAE and POLEMONIOIDEAE, the former containing *Cantua* and *Cobaea*, the latter the other ten genera. The genera and the number of their species are as follows: *Cantua* (6, with 2 new), *Cobaea* (9), *Polemonium* (29, with 3 new), *Collomia* (12), *Phlox* (49, with 9 new), *Gilia* (111, with 12 new), *Aliciella* (1), *Gymnosteris* (1), *Navarretia* (41, with 7 new), *Langloisia* (5, with 1 new), *Loeselia* (12), *Bonplandia* (1). The full synonymy and citation of collections, the elaborate splitting-up of the species into subspecies, varieties, and subvarieties, and the excellent illustrations, make the monograph a mine of information for students of the family. This careful traversing of American taxonomic work from the European standpoint is most instructive, and helps to focus attention upon our differences.—J. M. C.

**Tylostomeae.**—This “very natural tribe” of Gasteromycetes is described by LLOYD in a separately printed pamphlet,<sup>2</sup> which is dedicated to M. P. HARIOT, whose portrait forms its frontispiece. LLOYD recognizes seven genera, of which five are monotypic, *Tylostoma* containing the most of the species. All are illustrated by good photographs.—C. R. B.

### NOTES FOR STUDENTS

**Monograph of Ravenelia.**—A valuable contribution to the literature of the rusts has appeared in DIETEL'S<sup>3</sup> monograph of the genus *Ravenelia*. In the past, investigations relating to this peculiar genus have been necessarily of a more or less fragmentary nature, dealing with the morphology and taxonomy of a limited number of species only. Since the last monograph<sup>4</sup> which is at all comprehensive, a number of new species have been added to the genus from different parts of the world. On this account a full morphological and taxonomic treatment is especially desirable. The present paper is introduced by a discussion of

<sup>1</sup> ENGLER, A., *Das Pflanzenreich*. Heft 27. Polemoniaceae von A. Brand. pp. 203. figs. 39 (207). Leipzig: Wilhelm Engelmann. 1907. M. 10.20

<sup>2</sup> LLOYD, C. G., *The Tylostomeae*. 8vo. pp. 29. pls. 12. figs. 6. Cincinnati, O.: The author. 1906.

<sup>3</sup> DIETEL, P., *Monographie der Gattung Ravenelia* Berk. Beih. Bot. Centralbl. 20:343-413. pls. 5-6. 1906.

<sup>4</sup> DIETEL, P., *Die Gattung Ravenelia*. *Hedwigia* 33:22-29, 367-371. 1894.

the morphology of the genus. In general the mycelium, aecidiospores, and uredospores offer no striking characters different from other rusts; a few points of interest, however, are noted. The mycelium is usually intercellular and nourished by haustoria; but in some species it penetrates the cells, and in *R. atrocrustacea* on *Swartzia* it penetrates the vessels, developing abundantly within them. In several species the mycelium is perennial. Aecidia occur in only a few species and present no unusual peculiarities. The uredospores correspond to those of other rusts; but in the species on *Cassia* the uredospores are formed between the epidermal wall and the cuticle.

The teleutospores, which constitute the most interesting spore-type, are treated with some detail. A great many modifications exist as to the number of cysts, spores, and hyphae making up the spore-heads; but the author has arranged these into types within which the mode of building up the spore-head is remarkably uniform. These types are briefly as follows: (1) all spore-heads have a definite number of stalk-hyphae, which is uniform for each species, and each hypha gives a definite number of outer and inner spores; (2) the number of hyphae bearing the spore-heads is variable, but each hypha bears a definite number of spores, which is uniform within each species; (3) there is no uniformity in the number of hyphae and spores; the simplest forms of this last type have a single stalk-hypha which bears several spore-cells.

Heretofore the genus *Ravenelia* has been regarded as related to *Puccinia*, but on account of the occurrence of longitudinal divisions in the heads, DIETEL thinks the genus bears a closer relationship to the genera *Diorchidium*, *Anthomyces*, and *Sphaerophragmium*.

In the purely taxonomic part of the work 81 species are described, all of which are placed in the genus *Ravenelia*. The genera *Pleoravenelia* and *Neoravenelia*, recently separated from *Ravenelia* by LONG, are not retained, being considered insufficiently distinct. The author divides the genus into two sections: *Haploravenelia* and *Pleoravenelia*, the former comprising *Ravenelia* of LONG.—H. HASSELBRING.

**Bacteria and mineral salts.**—BENECKE,<sup>5</sup> in an investigation of the pigment-producing powers of *B. pyocyanus* and *B. fluorescens*, lays emphasis upon the need of careful purification of chemicals and selection of glass receptacles in bacteriological experiments with synthetic media. He used several kinds of glass control, including quartz, which is too expensive for general use, Jena glass which is K-free but contains Mg, resistance glass, Vienna glass which is probably Mg-free, Bohemian glass, and ordinary glass. With the various flasks he could obtain practically all the contradictory results of previous investigators, such as NOESSKE, THUM, JORDAN, GESSARD, SULLIVAN, and LOEW. By means of the controls he could trace most of the discrepancies in their results to impurities of chemicals or glassware. BENECKE's chemicals were washed and recrystallized

<sup>5</sup> BENECKE, W., Untersuchungen über den Bedarf der Bakterien an Mineralstoffen. *Botanische Zeitung* 65:1-23. 1907.

from 3-6 times in Jena glass or in platinum dishes; the water was redistilled and kept in Jena or in Thüringen glass. The most important of BENECKE's results, and those in which he is at variance with the investigators mentioned above, are as follows: (1) Dextrose or ammonium salts cannot be substituted for asparagin in the simple solution, asparagin 0.25 per cent., magnesium sulfate 0.05 per cent., potassium phosphate 0.02 per cent. (2) Potassium is necessary for development, although a very small amount of K-ions suffices for growth and pigment production. (3) Potassium cannot be replaced by lithium, sodium, or ammonium. It can be replaced by rubidium and caesium, though not to the same degree, the presence of 0.000015 per cent. of KCl being sufficient for development, while RbCl must be 10 times, and CsCl 100 times, as strong. The latter are also more toxic than KCl in concentrated solutions. (4) The presence of magnesium in the solution is necessary for development. Growth occurred in Mg-free solutions in Jena glass, which has 5 per cent. Mg in it, while no growth was obtained in Vienna glass. This Mg relation has been a stumbling-block for many investigators. On the other hand, the conclusion of most of the other authors that phosphate is essential in the solution is confirmed by BENECKE, as is also the observation of JORDAN and of NOESSKE that the acid group  $\text{SO}_4$  is necessary for pigment development in *B. fluorescens* and *B. pyocyaneus*. BENECKE, however, does not distinguish between the two pigments produced by the latter organism, which may not be alike dependent upon sulfate.—MARY HEFFERAN.

Infectious chlorosis.—Further studies of BAUR<sup>6</sup> on the infectious chlorosis of the Malvaceae have shown that green shoots which occasionally appear on variegated plants of *Abutilon Thompsoni*, Hort., are entirely immune to the disease and remain so. If a scion from one of these immune shoots is grafted upon a variegated plant, the scion produces only green leaves. If another scion of a green susceptible variety is grafted upon the first, the second scion will become variegated, showing that the virus is conducted unchanged through the immune piece. If, however, the experiment is conducted by using the immune species *Lavatera arborea* L. as the intermediate piece, no infection occurs in the second scion. In the first instance the intermediate plant is immune but does not affect the virus; in the second instance the virus is evidently destroyed. In his former paper BAUER showed that the virus causing chlorosis was developed only in the light. In the present paper the light relation is more fully investigated. It is found that somewhat shaded individuals, as when growing under shrubbery, lose their variegation, although growth is not greatly influenced. The optimum for virus formation, therefore, lies much above that for growth. Although shading the plants resulted in a diminution of the white areas, it was not possible by increased illumination to increase the areas above a certain maximum which

<sup>6</sup> BAUR, ERWIN, Weitere Mitteilungen über die infektiöse Chlorose der Malvaceen und über einige analoge Erscheinungen bei Ligustrum und Laburnum. Ber. Deutsch. Bot. Gesells. 24:416-428.

seems to be attained at ordinary illumination. This seems to indicate that even the green areas of infected leaves are permanently immune to the variegation. Both in blue and in red light plants retained their variegation.

BAUR has extended his investigations to other variegated plants and finds that infectious chlorosis is of widespread occurrence. Both true variegation and the infectious form may occur in varieties of the same species. Among the forms investigated, a variety of *Ligustrum vulgare* L. and one of *Laburnum vulgare* (Griseb.) owed their variegation to infectious chlorosis similar to that of *Abutilon Thompsoni*, Hort.—H. HASSELBRING.

**Hydrocyanic acid in plants.**—GUIGNARD has been giving special attention to plants which contain glucosides that produce this acid, and the conditions under which they may become poisonous. He relates in one paper<sup>7</sup> the cases of poisoning from the use of beans derived from wild or subspontaneous forms of *Phaseolus lunatus* L., a species having many forms, of which our Lima bean is one. In cultivation these are generally quite innocuous, but all contain phaseolunatin in greater or less quantity, which is split by emulsin into glucose, acetone, and hydrocyanic acid. The beans from Java have proved most virulent. A botanical and chemical history of the species is given, with illustrations of the seeds of many varieties. A new, convenient, and certain mode of detecting the acid is proposed. Filter paper is moistened with 1 per cent. picric acid and dried, then moistened with 10 per cent. sodium carbonate and dried, in which state it keeps its sensitiveness several months. A strip suspended in a test tube containing 0.02–0.05 mg HCN becomes red-orange in 12–24 hours.

In other papers<sup>8</sup> Guignard notes the presence of a cyanogenetic compound in *Sambucus nigra* L., *Ribes rubrum* L., *R. aureum* Pursh., where it is found in the fresh leaves from vigorous shoots, green fruits, and young bark of the stem, while traces appear elsewhere. The compound is probably a glucoside that is split by emulsin, which accompanies it and does not seem to be a reserve food.<sup>9</sup>

Emulsin he reports in the aerial roots of a considerable number of exotic and indigenous orchids,<sup>10</sup> and also in certain yeasts<sup>11</sup> (resembling *Sac. Pastorianus* Hansen). This has also been announced by HENRY and AULD.<sup>12</sup>

<sup>7</sup> GUIGNARD, L., Le haricot à acide cyanhydrique (*Phaseolus lunatus* L.). Revue de Viticulture 1906. col. pl. I. figs. 9.

<sup>8</sup> —, Sur l'existence, dans le sureau noir d'un composé fournissant de l'acide cyan-hydrique. Bull. Sci. Pharmacol. 12 and 13. 1905–6.

<sup>9</sup> —, Nouvelles observations sur la formation et les variations quantitatives du principe cyanhydrique de sureau noir. *Idem*. See also announcements in Compt. Rend. Acad. Sci. Paris June 3, July 24, Sept. 4, Dec. 26, 1905.

<sup>10</sup> —, Quelques faits relatifs à l'histoire de l'emulsine; existence générale de ce ferment chez l'Orchidées. *L. c.* footnote 8.

<sup>11</sup> —, Secretion d'emulsine par les levures. *L. c.*

<sup>12</sup> Proc. Roy. Soc. B. 76:568. 1905.

In the latest note<sup>13</sup> GUIGNARD adds a considerable number of Rosaceae, of the tribes Pruneae and Spiraeae, whose leaves and other parts furnish hydrocyanic acid.—C. R. B.

**Sexuality of the Uredineae.**—In 1904 BLACKMAN<sup>14</sup> showed that a peculiar process of fertilization occurs in theaecidium of *Phragmidium violaceum*, by which a nucleus from a vegetative cell of the mycelium migrates into a fertile cell, and thus brings about the condition of paired nuclei found by SAPPIN-TROUFFY<sup>15</sup> to be quite universal in the teleutospore-bearing mycelium. Two questions naturally arise as a result of this work. First, since theaecidium of *P. violaceum* is of a special type, how far will this process of fertilization be found to explain the origin of conjugate nuclei inaecidia generally? Second, what process takes place in those forms which have noaecidium? Both of these questions BLACKMAN and FRASER<sup>16</sup> attempt to solve in a later contribution to the cytology of the Uredineae. In *Uromyces Poae* Raben. and *U. Poarum* Neil., both of which are eu-forms with typicalaecidia, the migration of nuclei from one vegetative cell to another was observed in the tissue of theaecidium. These migrations were not so easily distinguished as in *P. violaceum*. In *Melampsora Rostrripi* Wagn., which hasaecidia of thecaeoma type, no fertilization was discovered, but there were some evidences that fertilization took place in the manner described by CHRISTMAN for *Phragmidium speciosum*. In *Puccinia Malvacearum* Mont., a lepto-form, the change from uninucleate to binucleate cells takes place in the hyphae of the teleutosorus, but the exact method could not be determined; neither could the transition be made out in the micro-forms *P. Adoxae*, D. C., *U. Scillarum* Wint., and *U. Ficariae* Lév.—H. HASSELBRING.

**The filiform apparatus.**—Striations on the micropylar portion of synergids were described in 1856 by SCHACHT, who called them "fertilization threads" (*Befruchtungsfäden*); HOFMEISTER gave the name "filiform apparatus" (*Fadenapparat*); STRASBURGER in 1882 believed the lines or threads consisted of fine pores. A paper by HABERMANN<sup>17</sup> now brings modern technic and modern lenses to bear upon the subject. The filiform apparatus, more or less developed, is characteristic of angiosperms generally. The apparatus arises by the transfor-

<sup>13</sup> ——, Nouveaux exemples de Rosacées à acide cyanhydrique. Compt. Rend. Acad. Sci. Paris 143:451. Oct. 1. 1906.

<sup>14</sup> BLACKMAN, V. H., On the fertilization, alternation of generations, and general cytology of the Uredineae. Annals of Botany 18:323-373. pls. 21-24. 1904.

<sup>15</sup> SAPPIN-TROUFFY, P., Recherches histologiques sur la famille des Uredinées. Le Botaniste 5: 59-244. figs. 68. 1896-7.

<sup>16</sup> BLACKMAN, V. H., and FRASER, MISS H. C. I., Further studies on the sexuality of the Uredinaceae. Annals of Botany 20:35-48. pls. 3-4. 1906.

<sup>17</sup> HABERMANN, ALFRED, Der Fadenapparat in den Synergiden der Angiospermen. Beih. Bot. Centralbl. 20:300-317. pl. 13. 1906.

mation of the foam-like cytoplasm, its growth continues by apposition, and the upper portion often swells considerably before fertilization. - The membrane of the embryo sac is resorbed over the apex of the synergids, which then quite frequently protrude. The vacuoles in the lower part of the synergids develop simultaneously with the filiform apparatus, and are separated from it by a plasma membrane. The filiform apparatus consists of cellulose, and its function seems to be the separation of the chemotactic glucose-containing substance, which passes into the micropyle and attracts the pollen tube. The name "synergid" is consequently quite appropriate.—CHARLES J. CHAMBERLAIN.

**Moisture in seeds.**—BROWN and DUVEL<sup>18</sup> have devised a method for the rapid determination of the percentage of moisture in grains. The method consists in heating a given weight of the grain in oil to drive off the water, which is condensed and measured in a graduated cylinder. The method is accurate, simple, and capable of great speed in application. The determinations are said to be accurate to 0.1 of 1 per cent., which makes it suitable for all scientific determinations. A determination can be made in twenty minutes, and with a number of compartments to the apparatus one trained manipulator with an ordinary assistant can make 200 determinations in a day. It is to be hoped that this will be one step toward putting grain-testing on a reliable basis. The economic improvement in the method of grading grain is seen when it is mentioned that the percentage of the European importation of corn that the United States furnishes has greatly diminished in the last decade, because of the great liability of our corn to spoil *en route*.—WM. CROCKER.

**Centrosomes in angiosperms.**—KOERNICKE<sup>19</sup> has reviewed the centrosome studies of the past seven years, paying particular attention to the work of BERNARD. Besides studying his old preparations and making new ones showing mitosis in the embryo sacs and pollen mother cells of various species of *Lilium*, KOERNICKE has made a thorough study of the division of the generative cell as it occurs in the pollen tube of *Lilium*, thinking that centrosomes might be retained longest by these structures which represent the ciliated sperms of some gymnosperms. No centrosomes were found, but on the contrary the fibers of the spindle were seen to end in the *Hautschicht*. It is of interest to note that while the generative cell is sharply outlined in the pollen grain, it loses its sharp contour after it passes into the pollen tube. After the generative nucleus divides, the two daughter nuclei lie free in the general cytoplasm of the pollen tube, there being no well-organized cells as figured by GUIGNARD and reproduced by current textbooks.—CHARLES J. CHAMBERLAIN.

<sup>18</sup> BROWN, EDGAR, and DUVEL, J. W. T., A quick method for the determination of moisture in grain. U. S. Dept. Agric., Bureau Pl. Ind., Bull. 99, pp. 24. figs. 12. 1907.

<sup>19</sup> KOERNICKE, MAX, Zentrosomen bei Angiospermen. Flora 96:501-522. pl. 5. 1906.

A birch rope.—D. P. PENHALLOW has described<sup>20</sup> a remarkable growth occurring upon a specimen of *Betula populifolia* found in the New Brunswick woods. It is a rope-like structure, reported to have been at least twenty feet long and approximately  $1 \times 0.4$  cm in diameter, and hanging free. The anatomical study showed that the growth had its origin in a lesion of the living bark. The traumatic reaction, instead of becoming localized and forming wood or sclerotic tissue, continued its development as active parenchyma and forced its way through the overlying and external tissue of the periderm. The form of the outgrowth suggests that it emerged through a lenticel. The outgrowth may be regarded as a special form of tumor, which developed by simultaneous division throughout its entire length and completed its growth in one season. The formation of cork proceeded over the entire surface until the final exhaustion of the parenchyma.—J. M. C.

Homology of the blepharoplast.—IKENO<sup>21</sup> replies to those who have questioned the correctness of the conclusion set forth in his paper on spermatogenesis in *Marchantia polymorpha*, and reasserts his belief that the blepharoplast is a centrosome. He admits, however, that the centrosome may be absent during the development of spermatogenous tissues in the higher liverworts, and may appear only as the blepharoplast. He thinks that the bodies now called blepharoplasts may not be homologous structures, and suggests three categories: (1) centrosome-blepharoplasts, which are either ontogenetically or phylogenetically of centrosome origin; here belong the blepharoplasts of myxomycetes, liverworts, pteridophytes, and gymnosperms; (2) plasmodesmal blepharoplasts, as in Chara and some Chlorophyceae; and (3) nuclear blepharoplasts, as in some genera of flagellates.—CHARLES J. CHAMBERLAIN.

The dwarf males of Oedogoniaceae.—A reinvestigation of this subject has brought PASCHER<sup>22</sup> to conclusions somewhat different from the commonly accepted views. The egg is regarded as a modified zoospore, the clear receptive spot corresponding to the hyaline ciliated area of the zoospore. The androzoospore (androspore of PRINGSHEIM) represents an intermediate development between the zoospore and spermatozoid. Accordingly the gynandrous and the macrandrous-dioecious forms of the Oedogoniaceae have advanced farther along the line of sexual differentiation than have the forms with dwarf males. The dwarf males of the Oedogoniaceae are homologous with the dwarf sporelings of the Chaetophoraceae and show more relationship with forms like Chaetophora than with forms like Ulothrix.—CHARLES J. CHAMBERLAIN.

<sup>20</sup> PENHALLOW, D. P., A birch rope; an account of a remarkable tumor growing upon the white birch. Trans. Roy. Soc. Canada II. 12:239-255. figs. 9. 1906.

<sup>21</sup> IKENO, S., Sur Frage nach der Homologie der Blepharoplasten. Flora 96: 538-542. 1906.

<sup>22</sup> PASCHER, ADOLF, Ueber die Zwergmännchen der Oedogoniaceen. Hedwigia 46:265-278. 1907.

**Secondary thickening in pteridophytes.**—The known cases of secondary thickening in recent Pteridophyta have been brought together by HILL<sup>23</sup> in a useful résumé. After stating the criteria for secondary growth, *Botrychium*, which has a distinct cambium, and *Ophioglossum*, which lacks a definite layer, are described, followed by *Angiopteris* and *Marattia*, in which a cambium forms a few xylem elements. CORMACK's observations on the secondary wood in the nodes of *Equisetum* are cited, though no reference is made to the cambium in the young cone as reported by JEFFREY.<sup>24</sup> The other cases of secondary growth include *Psilotum*, *Selaginella spinulosa*, and several species of *Isoetes*, especially *I. hystric*, which may show a cambium outside the vascular cylinder.—M. A. CHRYSLER.

**The cell wall.**—Whether isolated portions of protoplasm, without nuclei are capable of surrounding themselves with walls or not has long been in dispute. Nearly twenty years ago PALLA claimed that isolated portions of protoplasm could still form membranes. Later observers claimed that the non-nucleate fragments might still be connected with nucleated portions of the protoplasm by connecting fibers. Studies of the rhizoids of *Marchantia polymorpha* and the stinging hairs of *Urtica dioica* bring PALLA<sup>25</sup> to reassert his original view that non-nucleate portions of protoplasm can form membranes. He admits, however, that the portions of protoplasm must contain, as reserve substance, a material which can be used in building up a wall.—CHARLES J. CHAMBERLAIN.

**South African cycads.**—PEARSON<sup>26</sup> has begun the publication of a series of field notes on South African cycads, which promise to be of great interest. The first paper deals with *Encephalartos Friderici Guilielmi*, *E. Aliensteinii*, *E. villosus*, and an unnamed species of *Stangeria*. A summary of the observations is as follows: subterranean branching is a marked feature of the first-named species and of *Stangeria*; in the first two species of *Encephalartos* the strobili are lateral, occurring in cycles of three to six about the vegetative apex, which continues the growth of the stem; there is evidence that strobili are produced much more freely in exposed than in shaded situations; it is probable that entomophily occurs in *E. villosus*.—J. M. C.

**Apogamy in Elatostema.**<sup>27</sup>—TREUB adds *Elatostema acuminatum* (Urticaceae) to the increasing list of plants in which apogamy has been described.

<sup>23</sup> HILL, T. G., On secondary thickening in recent Pteridophyta. New Phytologist 5:208-214. 1906.

<sup>24</sup> JEFFREY, E. C., The development, structure, and affinities of the genus *Equisetum*. Mem. Boston Soc. Nat. Hist. 5:155-190. pls. 26-30. 1899.

<sup>25</sup> PALLA, E., Ueber Zellhautbildung kernloser Plasmateile. Ber. Deutsch. Bot. Gesells. 24:408-414. pl. 19. 1906.

<sup>26</sup> PEARSON, H. H. W., Notes on South African Cycads. I. Trans. S. African Phil. Soc. 16:341-354. pls. 6-8. 1906.

<sup>27</sup> TREUB, M., L'apogamie de l'*Elatostema acuminatum* Brogn. Ann. Jard. Bot. Buitenzorg 20:141-152. pls. 4-II. 1906.

The early stages in the development of megasporangia and also in the germination of the megaspore are normal; but a typical egg apparatus is not differentiated. The evidence that embryos are formed without fertilization is conclusive, but the evidence that some of the embryos may not come from nuclear tissue is not so conclusive. TREUB claims that the embryos come from contents of the embryo sac, but not from the egg, and consequently he uses the term apogamy rather than parthenogenesis.—CHARLES J. CHAMBERLAIN.

Anatomical classification of ferns.—FERNAND PELOURDE<sup>28</sup> has attempted to discover an anatomical basis of classification for the ferns. A great amount of detailed structure is described, and, like every other attempt to use one kind of character, the result is a readjustment of some of the old taxonomic lines. The genera represented in France are described upon this new basis, but its application to a wider range of forms remains to be demonstrated. As an addition of certain characters, to be used in connection with all other available characters, the research is a contribution; but as presenting a set of determining characters it can hardly be accepted at this late day.—J. M. C.

Nomenclature of desmids.—NORDSTEDT<sup>29</sup> has proposed the following rules of nomenclature for the desmids: (1) The nomenclature begins with RALFS, *British Desmidiae*, 1848; (2) The authors of names given earlier and accepted by RALFS must always be cited. It is also suggested that standard works for other groups of algae may be used in the same way, as for instance: HIRN, *Monographie und Iconographie der Oedogoniaceen*, 1900; BORNET et FLAHAULT, *Revision des Nostocacées hétérocystées contenues dans les principaux herbiers de France*, 1886-88; GOMONT, *Monographie des Oscillariées (Nostocacées homocystées)*, 1893.—J. M. C.

Chestnut disease.—In two additional papers MURRILL<sup>30</sup> emphasizes the unusual destructiveness of a disease of the American chestnut first made known by him. This disease is so virulent that it threatens to destroy all the chestnut trees in New York City and perhaps over large areas. The cause of the trouble is a species of Diaporthe newly described as *D. parasitica*. From observations and from experiments it is evident that the fungus attacks the trees only by the way of wounds. Attempts to infect young uninjured twigs and unfolding leaves gave negative results. No satisfactory treatment has as yet been found.—H. HASSELBRING.

<sup>28</sup> PELOURDE, FERNAND, Recherches anatomique sur la classification des fougères de France. Ann. Sci. Nat. Bot. IX. 4:261-372. figs. 80. 1906.

<sup>29</sup> NORDSTEDT, C. F. O., The starting point of the nomenclature of desmids. Botaniska Notiser 1906:97-118.

<sup>30</sup> MURRILL, W. A., A new chestnut disease. Torreya 6:186-189. 1906.

—, Further remarks on a serious chestnut disease. Jour. N. Y. Bot. Garden 7:203-211. figs. 25-30. 1906.

Bulbils and proembryo of *Lamprothamnus*.—Miss McNICOL<sup>31</sup> has been cultivating *Lamprothamnus alopecuroides*, one of the Characeae, for several years, the plants producing an abundant supply of antheridia and oogonia. Very few of the oospores seemed capable of germination, and proembryos produced from them resemble those produced from the underground nodes. For the most part, reproduction takes place by means of proembryos that are formed on the rhizoid-nodes and make use of the starch stored in the tubercles, which arise either directly or as transformed rhizoids.—J. M. C.

Semipermeable membrane of grass seeds.—BROWN<sup>32</sup> has published a preliminary account of a semipermeable membrane which incloses the grain of *Hordeum*, which permits the passage of water and iodin, but prevents the passage of sulfuric and hydrochloric acids and all salts of metals examined, when in aqueous solution. The semipermeable property is not due to the action of living protoplasm and is located in the "spermoderm" of the grain. The same covering occurs in grains of *Avena*, *Triticum*, and *Secale*. If these observations are correct, the "spermoderm" is a most remarkable structure.—J. M. C.

Synapsis and synizesis.—Following McCLEUNG, SCHAFFNER<sup>33</sup> uses the term synizesis to describe the familiar contracted condition of the chromatin usually described as synapsis; while he reserves the term synapsis in a very restricted sense as applying to the chromatin fusions which take place during the contracted state. The synizetic knot is not always around or in contact with the nucleolus, and while usually lateral it is often central in position. SCHAFFNER'S figures show that the synizetic knot has no relation to gravity.—CHARLES J. CHAMBERLAIN.

Excentric growth.—URSPRUNG<sup>34</sup> records experiments calculated to throw some light on the cause of excentric growth of stems. Seedlings of *Ricinus* fixed in a horizontal position showed an excentric growth of the hypocotyl, with mechanical cells in the upper part and large thin-walled cells in the lower part. The reason for these appearances is not clear, for it has been shown that mere tension, such as would result in the present instance from the turgescence of the cells on the lower side, does not result in the production of mechanical tissue.—M. A. CHRYSLER.

Growth of lignified cell walls.—It is usually assumed that lignified cell walls, such as those of tracheids, are incapable of further growth. But this idea

<sup>31</sup> McNICOL, MARY, The bulbils and proembryo of *Lamprothamnus alopecuroides* A. Braun. Annals of Botany 21:61-70. pl. 8. 1907.

<sup>32</sup> BROWN, A. J., On the existence of a semipermeable membrane enclosing the seeds of some of the Gramineae. Annals of Botany 21:79-87. 1907.

<sup>33</sup> SCHAFFNER, J. H., Synapsis and synizesis. Ohio Naturalist 7:41-48. pl. 4. 1907.

<sup>34</sup> URSPRUNG, A., Beitrag zur Erklärung des exzentrischen Dickenwachstums an Krautpflanzen. Ber. Deutsch. Bot. Gesells. 24:498-501. 1906.

is negatived by the observation of URSPRUNG<sup>35</sup> that the pith of such plants as *Sambucus* and *Tectona* increases considerably in diameter after the wood cylinder is formed. This can be brought about only by expansion and division of the elements of the wood. The author holds that vessels can increase in diameter after they have lost their living contents.—M. A. CHRYSLER.

Waterbloom.—MÖBIUS<sup>36</sup> reports that for several years a waterbloom has appeared each summer in the botanical garden in Frankfurt, and that it is regularly composed of three species of Cyanophyceae: *Oscillatoria Agardhii* Gomont, *Anabaena flos-aquae* Bréb., and *Clathrocystis aeruginosa* Henfrey. The appearance of *Oscillatoria* in the association has not been noted hitherto. An abnormal form of *Cladophora crispata* (Roth.) Kütz. is also described.—CHARLES J. CHAMBERLAIN.

Abnormal mosses.—Two interesting abnormalities are described by GYÖRFFY,<sup>37</sup> collected in the High Tatra. A specimen of *Plagiomnium demissum* Lindb. shows the seta forking at the apex and carrying two perfect capsules; and one of *Polytrichum alpinum* L. has two setae, each carrying a normal capsule, both covered by a single calyptra. BRUCH records a like case in *P. juniperinum*. Obviously these forms have arisen from a single egg.—C. R. B.

Respiration.—KOSTYTSCHEW declares that in the aerobic respiration of the leaves of seed plants which contain mannit, hydrogen is produced; but in anaerobic respiration, even when very vigorous, not a trace is set free.<sup>38</sup> The experiments were designed to test the results of MUNTZ (1876) and DELUCA (1878), which antedated bacteriological knowledge, that H is produced by mannitiferous fungi and seed plants in anaerobic respiration.—C. R. B.

Anatomy of Urticaceae.—RENNER<sup>39</sup> has published a detailed account of the anatomy of Artocarpeae and Conocephaleae, paying special attention to *Ficus*. A systematic presentation of the tribes follows, the anatomical structure being used as a basis of discussion.—J. M. C.

<sup>35</sup> URSPRUNG, A., Ueber die Dauer des primären Dichenwachstums. Ber. Deutsch. Bot. Gesells. 24:489-497. 1906.

<sup>36</sup> MÖBIUS, M., Algologische Beobachtungen über eine Wasserblüthe und eine Cladophora. Hedwigia 46:279-287. 1907.

<sup>37</sup> GYÖRFFY, I., Bryologische Beiträge zur Flora der Hohen Tatra. IV. Hedwigia 46:262. 1907.

<sup>38</sup> KOSTYTSCHEW, S., Zur Frage über die Wasserstoffausscheidung bei der Atmung der Samenpflanzen. Ber. Deutsch. Bot. Gesells. 24:436-441. 1906.

<sup>39</sup> RENNER, OTTO, Beiträge zur Anatomie und Systematik der Artocarpen und Conocephaleen, insbesondere der Gattung *Ficus*. Bot. Jarb. 39:319-448. 1907.

## NEWS

DR. JOHN W. HARSHBURGER has been promoted to be assistant professor of botany at the University of Pennsylvania.

INFORMATION has come from Kew that in its future publications the nomenclature will be in accord with the rules adopted by the Vienna Congress.

DR. J. C. ARTHUR, Purdue University, gave an illustrated lecture March 30 in the Field Museum (Chicago) Spring lecture course, his subject being "A superposed vegetation—the plant rusts."

DR. EDWIN B. COPELAND returned from Manila about the middle of March and has taken up research work in plant physiology at the Agricultural Experiment Station of the University of West Virginia, at Morgantown.

THE HOUSE of PAUL PAREY in Berlin has recently issued five new plates of the well-known series by Dr. L. KNY. The new plates are the first of a second hundred, which are to be increased in size from  $67 \times 82\text{cm}$  to  $106 \times 150\text{cm}$ . They illustrate Drosera, Mimosa, Spirogyra, Cuscuta, and Berberis.

OTTO KUNTZE died suddenly at San Remo on January 28, at the age of 64. His earliest work as a taxonomist appeared in 1867; but since the appearance of his *Revisio Generum* (1891-98) he has been known chiefly for his writings on nomenclature. The *Journal of Botany* justly says: "It is in connection with the revision of nomenclature that his name will chiefly be remembered, and it is to be regretted that the intolerance of his views and the intemperance of the language in which they were stated led to a somewhat insufficient appreciation of his labor and research."

THE UNIVERSITY OF MINNESOTA has recently completed, at a cost of \$10,000, a range of plant houses covering about 370 square meters, and consisting of five connected houses and a work room ( $5 \times 7.5\text{m}$ ) with basement for heating apparatus. There is a xerophyte house ( $7.5 \times 7.5\text{m}$ ), palm house ( $8.5 \times 10 \times 5.5\text{m}$  high), aquatic house ( $5.5 \times 10\text{m}$ ), lily house ( $5.5 \times 10\text{m}$ ), and a temperate house. An additional sum of \$7000 has been asked to build a laboratory greenhouse connected with the others and to improve the grounds as a botanic garden. Unfortunately the plant houses and grounds are widely separated from the laboratories.

AT THE thirty-seventh annual meeting of the Wisconsin Academy of Sciences, Arts, and Letters held at Madison, Wisconsin, February 7 and 8, 1907, the following botanical papers were presented: E. W. OLIVE: Nuclear migrations and cell fusions in the rusts; A. H. CHRISTMAN: The morphology of the spore forms of the rusts; R. A. HARPER: Heredity in the lower fungi; J. B. OVERTON: Diakinesis in Thalictrum; W. G. MARQUETTE: Concerning the organization of the cell in Marsilia; B. F. LUTMAN: Cell and nuclear fusions in the promycelial cells of certain smuts; C. E. ALLEN: The distribution of grandparental characters in Pisum.

# BOTANICAL GAZETTE

MAY 1907

## CONSPECTUS GENERIS AMORPHAE

CAMILLO KARL SCHNEIDER

In reviewing the genus *Amorpha* for my *Illustriertes Handbuch der Laubholzkunde*, I had the opportunity of looking over the forms of this small but variable genus, for Professor TRELEASE had kindly sent me the rich collection of the Herbarium of the Missouri Botanical Garden. I am very much indebted to him for his great assistance to me in all my dendrological studies.

In SMALL'S *Flora S. E. United States*, F. E. BOYNTON has published a good synopsis of most of the species of the genus, but there seem to me to be some errors of statement arising from a failure to clear up some of the descriptions of the older European and American authors. I take this opportunity, therefore, to refer to some points concerning which I think BOYNTON has taken a wrong view.

### CLAVIS ANALYTICA SPECIERUM

1a. Foliola infima proxime super basin subdilatatum petiola adnata ramo valde approximata fere pseudostipulosa. 2.

1b. Foliola infima distincte paulo superius rhachi adnata a ramo plus minusve distantia. 3.

2a. Calycis dentes omnes fere aequilongi, tubo paullo breviores; foliola 15-51, subsessilia, ovato-oblonga vel oblongo-elliptica, basi rotunda, apice fere semper subacuta, valde approximata; planta omnibus partibus albo-canescens

2. *A. canescens*

2b. Calycis dentes diversi, superiores breviores et paullo obtusiores (sed satis variabiles); foliola distinctius petiolulata, elliptica vel anguste-elliptica, basi semper apice plerumque rotunda; planta haud distincte denseque canescens sed plus minus pubescens; interdum subglabrescens . . . 1. *A. herbacea*

3a. Folia minus quam 10<sup>cm</sup> longa; foliola 15-35, brevepetiolulata, anguste-elliptica vel elliptica, utrinque rotunda vel subrotunda, 6-13<sup>mm</sup> longa et 2.5-7<sup>mm</sup>

lata; calycis dentes omnia subacuminata, superiores paullo breviores, inferiores tubo fere subaequilongi; petala obovata, apice leviter emarginata, basi in unguem brevissimum contracta; fructus dorso fere recti nec non recurvati 4-5<sup>mm</sup> longi, glanduloso-verrucosi; planta adulta omnibus partibus glabriuscula . . . . . 3. *A. microphylla*

3b. Folia longiora; foliola majora vel planta ceteris signis diversa. 4.

4a. Foliorum rhaches ramulique juniores glanduloso-hispiduli (*i. e.* glandulis pedicellatis aculeiformibus sparsis obtecti); calycis dentes subacuminati, subaequilongi, tubo fere paullo longiores . . . . . 5. *A. hispidula*

4b. Glandulae pedicellatae semper deficientes. 5.

5a. Foliola pro genere permagna, vix minus quam 3.5-4<sup>cm</sup> longa, pagina inferiora nervis lateralibus distincte elevatis, canescens, subcoriacea; planta ramis junioribus rhachibus foliorum et inflorescentiarum dense tomentosa . . . . . 14. *A. paniculata*

5b. Nervi laterales foliorum subitus non vel vix elevati vel planta partibus indicatis haud tomentosa. 6.

6a. Calycis dentes brevissimi et obtusissimi, multo latiores quam longi; petala fere orbicularia, basi brevissime unguiculata; folia ad 20<sup>cm</sup> longa; foliola 9-19, ovata, ovato-elliptica vel late elliptica, basi rotunda vel subrotunda, apice obtusa vel leviter emarginata, terminalis ad 6×3<sup>cm</sup> magna; fructus dorso recti, 7-8<sup>mm</sup> longi, sparse glanduloso-verrucosi; planta fere omnibus partibus glaberrima . . . . . 7. *A. glabra*

6b. Calycis dentes semper distincti, acuti vel planta ceteris signis diversa. 7.

7a. Calycis dentes omnes fere aequilongi distincte acuminati, tubo sublongiores; folia 10-15<sup>cm</sup> longa; foliola 13-21, valde approximata, marginibus se integentia, ovato-elliptica vel elliptica, basi rotunda, apice rotundata vel obtusa, vix emarginata, mucrone brevi instructa, 2.5-3.6<sup>cm</sup> longa et 1.2-1.7<sup>cm</sup> lata, brevi-petiolumata, nervis lateralibus subtus subelevatis; fructus (an maturi?) dorso, ut videtur, recti, 0.5<sup>mm</sup> longi, glanduloso-verrucosi; planta omnibus partibus subtomentella . . . . . 4. *A. Schwerini*

7b. Calycis dentes diversi, superiores breviores et nondum acuminati vel foliola semper conspicue remota, marginibus inter se non contingentia. 8.

8a. Folia 7-16<sup>cm</sup> longa; foliola 10-33, elliptica vel anguste-elliptica, utrinque rotundata, pleraque levissime emarginata, mucronulata, breve sed distincte petiolulata, 7×5<sup>mm</sup> ad 17×7<sup>mm</sup> vel 15×5<sup>mm</sup> magna, fere subcoriacea; dentes calycis superiores subacuti, inferiores subacuminati longiores; fructus dorso recti, 0.5<sup>mm</sup> longi, glanduloso-verrucosi; planta omnibus partibus glabriuscula sed inflorescentia distinctius pubescentibus . . . . . 8. *A. caroliniana*

8b. Foliola majora vel planta ceteris signis diversa. 9.

9a. Foliola satis magna, vix minus quam 1.5<sup>cm</sup> lata, basi rotunda vel cordata, subcoriacea; fructus dorso fere recti vel ad apicem subito leviter recurvati. 10.

9b. Foliola distincte angustiora vel basi subacuta vel tenuiora membranacea vel fructus dorso manifeste recurvata (*falcata!*). 11.

10a. Foliola superne nitentia, plus minus duplo longiora quam lata, 2.2-4×

1. 4-2<sup>cm</sup> vel ad 6×2.5<sup>cm</sup> magna; petala obovato-cordata, brevi-unguiculata, 6-7<sup>mm</sup> longa; fructus maturi 7-9<sup>mm</sup> longi; dorso ad apicem subito leviter recurvati, dense vel vix glanduloso-verrucosi . . . 12. *A. virgata* (et *A. nitens*)

12a. Foliolo superne haud nitentia, minus quam duplo longiora quam lata, 0.5×1.2 vel 2×1.5 ad 5×4 vel 5.5×3.5<sup>cm</sup> magna; petala basi in unguem distinctum contracta; fructus dorso recti, 7<sup>mm</sup> longi, glanduloso-verrucosi; planta omnibus partibus pubescentia vel glabriuscula

13. *A. laevigata* (et *A. texana*)

13a. Folia 9-20<sup>cm</sup> longa; foliola 11-17, ovato-elliptica vel elliptica, basi late subacuta, raro manifeste rotundata, apice obtusa vel rotunda vel subacuta, 2.3-4×1.2-2.2<sup>cm</sup> magna; petiolula ad 3<sup>mm</sup> longa; petala obovato-oblonga, 0.5<sup>mm</sup> longa; fructus 0.6<sup>mm</sup> longi, vix (an semper?) glanduloso-verrucosi; planta adulta glabriuscula . . . . . 6. *A. californica*

13b. Planta foliis vel fructibus vel signis ceteris diversa. 12.

12a. Fructus dorso fere recti plus minus distinete, 6<sup>mm</sup> longi, glanduloso-verrucosi; folia 8-15<sup>cm</sup> longa, foliola 12-31, anguste elliptica vel anguste ovato-lanceolata, utrinque rotundata, apice mucronulata, 12-18×6-8<sup>mm</sup> magna, brevi-petiolulata; dentes calycis breves, superiores fere rotundi, inferiores acuti medio paullo longiore; petala obovata, brevi-unguiculata, 0.5<sup>mm</sup> longa; planta omnibus partibus junioribus molliter pubescens, deinde glabriuscula

9. *A. tennesseensis* (cf. etiam 10)

12b. Fructus manifeste falcata (dorso recurvata), fere semper longiora vel foliola latiora vel utrinque acuta. 13.

13a. Folia 9-20<sup>cm</sup> longa; foliola 15-27, lanceolata, utrinque (praecipue basi) acuta vel apice subrotundata, mucronulata, 2-3.2×0.7-0.8 vel rarius ad 3×1.2-1.5<sup>cm</sup> magna, distinete petiolulata; fructus maturi 6-7.5<sup>mm</sup> longi; planta adulta satis glabriuscula, sed folia non glabra

10. *A. angustifolia* (cf. etiam 9)

13b. Folia ad 30<sup>cm</sup> longa; foliola 11-25, plerumque majora, forma variabilis; fructus maturi 8-9<sup>mm</sup> longi; planta pubescens vel adulta plerumque glabriuscula . . . . . 11. *A. fruticosa*

In the following pages I shall comment upon each species and its forms and shall attempt to clear up its synonymy. It also seems best to cite all the specimens I have seen. Figures of most of the details of leaves, calyx, petals, and fruits will be found in my *Illustriertes Handbuch* 2:pt. 1. 1907.

1. *A. HERBACEA* Walt. Fl. Carol. 179. 1788.—*A. pubescens* Willd. Berlin. Baumzucht 17. 1796; *A. pumila* Michx. Fl. Bor.-Am. 2:64. 1803.

Var. *a. TYPICA*.—Foliis majoribus, ad 25<sup>cm</sup> longis; foliolis 15-37-35, ad 2.5×1.3<sup>cm</sup> magnis.

NORTH CAROLINA.—Bladen Co., *Bilm. Herb.* no. 35, 9. ix. 96 and no. 35b, 20. vi. 97; Buncombe Co., *Bilm. Herb.* no. 35a, 9. vii. 98.

SOUTH CAROLINA.—Aiken Co., *H. Eggert*, 24. iii. 99.

GEORGIA.—Richmond Co., *H. Eggert*, 22. v. 99.

FLORIDA.—Sumter Co., *Curtiss*, no. 5414; *Tracy*, no. 6831 (locality unreadable).

Var. *β. Boyntoni*, var. nov.—Differt: foliis minoribus, ad 18<sup>cm</sup> longis; foliolis valde approximatis, 33–70, tantum ad 18×6–7<sup>mm</sup> magnis.

GEORGIA.—Bulloch Co., *R. M. Harper*, no. 942.

FLORIDA.—Lake Co., *Hitchcock*, in the vicinity of Eustis, vi–vii. 94; *G. V. Nash*, no. 1147; Pasco Co., *Curtiss*, no. 6664; Hernando Co., *Hitchcock*, vi–vii. 98; ad fl. St. John's *Rugel*, no. 166; ad ripas fl. Manatee, *Rugel*, no. 165.

A very graceful variety, with its numerous small leaflets, but apparently connected with the type by some intermediate forms, so that I do not venture to treat it as a species.

2. A. CANESCENS Nutt., *Fraser Cat.* 1813.

Var. *a. TYPICA*.—Foliolis apice plus minus distincte acuta vel subacuta, etiam adulta subitus cinereocani; inflorescentiis plerumque 12–15<sup>cm</sup> longis.—Vidi specimina numerosissima ex regionibus subtus indicatis.

Saskatchewan, Dakota, Minnesota, Wisconsin, Nebraska, Iowa, Illinois, Missouri, Arkansas, Indian Territory, Texas (*Lindheimer*, vii. 42, dry prairies, Lynchburg), New Mexico (*Earle*, no. 205 ex parte, El Capitan Mts.; *Brandegee*, no. 12023, San Miguel Co.).

Var. *β. GLABRATA* Gray, *Pl. Wright*. 1:49. 1852. emend. (incl. var. *leptostachya* Engelm. apud Torr. and Gray, *Pl. Fendl.* 31. 1849. nom. nud. [sec. specim. orig.!]).—Differt: foliolis obtusioribus, utrimque fere aequaliter rotundatis, supra glabrescentia, subtus glabriuscula, viridescentia; inflorescentiis plerisque longioribus (ad 30<sup>cm</sup>).

NEW MEXICO.—San Miguel Co., *Fendler*, no. 125.

MISSOURI.—Franklin Co., *Herb. Fritchey*, 16. vi. 86; *Bush*, no. 735. Ewan (?); *Dewart*, no. 69, Springfield.

This form is apparently connected with the type by the following specimens; *Earle*, no. 205 ex parte (New Mexico); *Lüders*, 1842, Franklin Co., Missouri; *Mead*, 1843, Augusta, Illinois.

A glabrescent form with very acute leaflets was collected by *Lapham*, 1843, Milwaukee, Wisconsin.

There is another remarkable form of the type with very large and obtuse leaflets ( $ad 2 \times 1.3$ <sup>cm</sup> magnis) of the lower leaves, from Watson, Missouri, *Bush*, no. 217.

3. A. *MICROPHYLLA* Pursh, Fl. Am. Sept. 2:466. 1814.—*A. nana* Nutt. Gen. N. Am. Pl. 2:91. 1818; nec Nutt. 1813 (*fide* Torr. et Gray, Fl. N. Am. 1:690. 1840) non Sims 1820.

Vidi specimina numerosa ex Manitoba, Dakota, Minnesota, Iowa, Nebraska, et New Mexico (*Gordon*, no. 13, iv. 48, Upper Canadian River).

4. A. *Schwerini*, sp. nov.

NORTH CAROLINA.—Rowan Co., *J. K. Small*, 18-27. viii. 94, on Dunn's Mt. (fructibus semi-maturis).

With its very long acuminate calyx teeth, and its very approximate, rather large leaflets pubescent on both sides, this form seems to be a good but very local species.

5. A. *HISPIDULA* Greene, Fl. Francisc. 1:14. 1891.—*A. californica* Hook. et Arn. Bot. Beech. Voy. 333. 1841, nec Nuttall apud Torr. et Gray.

CALIFORNIA.—Shasta Co., *H. E. Brown*, no. 232½, 15-18. v. 97; Ventura Co., *Elmer*, no. 3950; Monterey Co., *Elmer*, no. 3280; Sonoma Co., *Heller*, no. 5757; Los Angeles Co., *LeRoy Abrams*, no. 2622; *H. E. Hasse*, v. 92; Orange Co., *LeRoy Abrams*, no. 1828; San Bernardino Co., *G. Engelmann*, 21. ix. 80; *Parry* et *Lemmon*, no. 10811 (no. 97); *Parish*, nos. 3198 et 4185; *H. M. Hall*, no. 1288.

A very distinct species with its prickle-like glands. HOOKER and ARNOTT say (*l. c.*) "ramulis petiolisque glandulis rigidis acutis *aculeatis*;" while NUTTALL (apud Torr. et Gray) makes no mention of such glands and plainly says "teeth of the villous calyx all acute and *short*;" but it is somewhat strange that he also says "petioles furnished with *minute glandular scales*." Unfortunately I had no opportunity to see an original specimen of Nuttall from Santa Barbara, where *A. hispidula* seems to occur very frequently.

6. A. *CALIFORNICA* Nutt. apud Torr. et Gray, Fl. N. Am. 1:306. 1838.

CALIFORNIA (southern).—San Francisco Mts., *Hall*, no. 2121; San Diego Co., *Orcutt*, near Jacumba valley, vi. 90; *LeRoy Abrams*, no. 3425; *Cleveland*, no. 1361, in 1874.

ARIZONA.—Santa Rita Mts., *Pringle*, 5. vi. 84; *Engelmann*, 27. ix. 80 (in the leaflets this specimen very much resembles *A. angustijolia*, but the rather glandless small fruits are quite the same as those of the Mexican plant); Fort Whipple, *E. Palmer*, no. 240.

NEW MEXICO.—Grant Co., *Metcalfe*, no. 133; Albuquerque, *Harward*, no. 12; Doña Ana Co., *Wooton*, no. 46.

MEXICO.—Chihuahua, *Pringle*, no. 1588 (Paso del Norte).

This species seems very nearly allied to *A. fruticosa* (and also to *A. angustifolia*), but the fruits are smaller and only a little falcate. Unfortunately I saw only very few mature fruits, and I believe that the following forms may belong to *A. californica* rather than to *A. fruticosa*; some of them probably represent *A. laevigata* Boynton, but I never found a standard broader than long. For further comments see under *A. laevigata*.

TEXAS.—Lynchburg, *Lindheimer*, vii. 42; Comale Creek, *Lindheimer*, no. 335, vi. 45; without locality, *Lindheimer*, no. 38, in 1843; Gillespie Co., *Jermy*; Comanche Co., *Eggert*, 8. v. 00; Dallas Co., *Eggert*, 23. vi. 99 (the plant of 24. vi seems to be *A. angustifolia*).

And with (?) the following:

ARKANSAS.—Fayetteville, *Harvey*, no. 19.

COLORADO.—Fort Collins, *Crandall*, 12. vi. 96; Denver, *Bisson*, no. 13a; Military Park, *E. C. Smith*, 26. vi. 91.

7. *A. GLABRA* Poiret, in Lam. *Encycl. Suppl.* 1:330. 1810, nec Boynton.—?*A. glabra* Desf. *Cat. Hort. Paris* 192. 1804 (*nomen nudum*) et Persoon, *Syn.* 2:295. 1807 (*nomen nudum*); *A. montana* Boynton, *Biltm. Bot. Stud.* 1:138. 1902.

NORTH CAROLINA (western).—Rutherford Co., *Rugel*, v. 41, in montibus ad Broad River; *Biltm. Herb.*, no. 14f, 10. v. 98; Polk Co., *Biltm. Herb.*, no. 14d, 31. v. 97; Biltmore, *Biltm. Herb.*, no. 14, 13. v et 29. viii. 96 et no. 14b, 2. vi. et 20. ix. 97.

TENNESSEE (eastern).—Knoxville, *A. Ruth.* no. 304.

POIRET (*l. c.*) gives a very clear description in French and the following short but satisfactory diagnosis in Latin: "Amorpha glabra, foliolis pedicellatis, obtusis; dentibus calycinis omnibus obtusis, brevissimis." BOYNTON and all other authors seem to have completely overlooked POIRET's description. *A. glabra* also seems to be a very local species, like *A. Schwerini*.

8. *A. CAROLINIANA* Croom, *Am. Jour. Sci.* 25:74. 1834 (confer etiam apud Torr. et Gray, *Fl. N. Am.* 1:305. 1838).—*A. cyano-stachya* Curtis, *Jour. Boston Soc. Nat. Hist.* 1:140. 1835 (1837), vidi specim. orig.; *A. glabra* Boynton, in *Small Fl. S. E. United States* 626. 1903.

NORTH CAROLINA (eastern).—Newborn, *Loomis* (*fide* Croom; spec. orig. non vidi!); Wilmington, *Curtiss* (vidi!).

GEORGIA.—Johnson Co., *R. M. Harper*, no. 1347; very similar also Lloyd Co., *Chapman Herb.*, no. 1322 (or 1522).

FLORIDA.—*Biltm. Herb.*, no. 5767; *Herb. Chapman* (this specimen has been named by BOYNTON *A. glabra* Desf.); there are also in *Herb. Mo. Bot. Garden* two sheets of *Herb. Chapman* no. 1524 without any other indication.

TEXAS.—Very similar are some small specimens from Columbia, *Bush*, nos. 962 and 1581.

*A. caroliniana*, as I understand it, is a very glabrous species, with leaflets relatively shorter and broader than those of *A. tennesseensis* and *A. angustijolia*; but without very ripe fruits it is often difficult to say to what species such a form may belong. Only specimens collected in September have the fruits fully ripened.

9. *A. TENNESSENSIS* Shuttlw. in *Kunze Delect. Sem. Hort. Lips.* p. 1. adn. 1848, et in *Linnaea* 24:191. 1851 (descript. non sufficiens); vide Boynton in *Small Fl. S. E. United States* 625. 1903.

TENNESSEE.—Prope Dandridge, *Rugel*, vi et ix. 42 (specim. orig., fructibus maturis dorso fere rectis!); Polk Co., *Biltm. Herb.*, no. 1381a.

It is not without some reservation that the following specimens are placed with this species:

NORTH CAROLINA.—Stanley Co., *Small*, 18. viii. 92.

FLORIDA.—John's Pass, *Tracy*, no. 7794.

ALABAMA.—Colliert Co., *Eggert*, 21. v. 99, Tennessee River near South Florence (foliis infimis cum foliolis ad  $3 \times 1.5^{\text{cm}}$  magnis!).

LOUISIANA.—Port Eads, *Tracy et Lloyd*, no. 176.

ARKANSAS.—Bertig, *Trelease*, 28. x. 97; Beaver Sta., *Glatfeller*, 18. vii. 98; *Herb. Engelmann*, no. 298, July 1835.

MISSOURI.—Jasper Co., *Trelease*, nos. 195 et 201. (in 1897).

ILLINOIS (southern).—Elsah, *Reed*, vi. 98. (an *A. fruticosa* var. *humilis* [Tausch] m.?).

10. *A. ANGUSTIFOLIA* Boynton, *Biltm. Bot. Stud.* 1:139. 1902.—*A. fruticosa* var. *subglabra* Gray, *Jour. Bost. Soc. Nat. Hist.* 6:174. 1850.

A somewhat doubtful species, apparently connected with *A. fruticosa* (and probably also with *A. californica*) by intermediate forms. BOYNTON cites as synonym *A. fruticosa* var. *angustifolia* Pursh, *Fl. Am.-Sept.* 2:466. 1814, but I think it is very difficult to decide what form PURSH named var. *angustifolia* without having at hand his original specimens. He gives no other locality of his *A. fruticosa* than "Carolina and Florida." Concerning his *A. angustifolia* he remarks, "v. s. in *Herb. Lewis*." Probably the var. *angustifolia* of Pursh is the same as *A. humilis* Tausch, which is only a small form of typical *A. fruticosa*. The typical *A. angustifolia* according to BOYNTON has elliptic to linear-oblong leaflets which are distinctly acute at both ends. The fruits are as falcate as in *A. fruticosa*. In habit it very much resembles *A. tennesseensis*, but that species has more numerous and more approximate leaflets, which are rounded or obtuse at each end. The following specimens seem to me typical *A. angustifolia*:

TEXAS.—New Braunfels, *Lindheimer*, no. 595; Kerr Co., *Heller*, no. 1596; Dallas Co., *Eggert*, 24. vi. 99 (cf. etiam sub *A. californica*); Mitchell Co., *Eggert*, 8. vi. 90.

INDIAN TERRITORY.—Sapulpa, *Bush*, nos. 70 et 1105.

NEW MEXICO.—Pawnee Fork, *Fendler*, no. 126.

KANSAS.—Manhattan, *Norton*, 9. viii. 92, and also Riley Co., no. 89 (and also no. 89a?).

IOWA.—Steamboat Rock, *Pammel*, *Hume*, and *Fitz*, no. 1621.

The following specimens seem to be somewhat intermediate between *A. angustijolia* and *A. fruticosa* (especially its var. *humilis*, which can be determined clearly only from living specimens!).

MISSOURI.—Taney Co., Forsyth, *Wm. Trelease*, 7. viii. 97.

COLORADO.—Beaver Co., *Redfield*, no. 71 (vel 1365).

SOUTH DAKOTA.—Lake Campbell, *Herb. Luke*, 13. vi. 91.

WYOMING.—Laramie Co., *Nelson*, no. 8657 (forma foliolorum valde variabilis!).

II. *A. FRUTICOSA* L., Sp. Pl. 2:713. 1753.—*A. perforata* Schkuhr, Bot. Handb. 2:333. 1808; *A. elata* Hayne, Dendrol. Fl. 134. 1822; *A. pubescens* Schlecht. Linnaea 34:691. 1851.

The type of this species is very clearly indicated by Linnaeus in citing "Hort Cliff, p. 353. tab. 19," but I have seen only a relatively small number of specimens which represent this type. It was collected first by *Catesby* in North Carolina. I distinguish the following varieties, but I believe that such widely distributed forms as *A. angustijolia* and *A. tennesseensis* can only be taken for varieties of *A. fruticosa*. Probably also *A. californica* and *A. caroliniana* may be considered as mere geographical varieties of *A. fruticosa*.

Var. *TYPICA*.—Frutex elata partibus omnibus junioribus sericeo-pubescentibus adultis glabriusculis vel glabris.

NORTH CAROLINA.—Stanley Co., *Small* et *Heller*, no. 380 (a form somewhat intermediate between typical *A. fruticosa* and *A. tennesseensis*); loco non indicato, *S. B. Buckley*.

ILLINOIS.—Stark Co., *V. H. Chase*, no. 737; Washington Co., *French*, no. 1362 (forma pube var. *croceolanata* similis).

IOWA.—Ames, *Ball* et *Meeker*, no. 525, et *Hitchcock*; Story City, *Pammel* et *Ball*, no. 1753 (?), 4. vi. 98; Decatur Co., *Anderson*, 9. vi. 01.

MISSOURI.—Jefferson Co., *Eggert*, 5. v. 96; *Dr. O. Krause*, 23. v. 66; St. Louis Co., *Eggert*, 6. v. 75; Webb City, *Bush*, no. 553; Bridgeton, 28. v. 59; Jackson Co., *Bush*, 13. vi. 92.

NEBRASKA.—Saye Creek, *Hayden*, 22. vi. 53; Kearney, *Pammel* et *Brownlee* (?), 27. vi. 99.

KANSAS.—Cowley Co., *Madwhite* (?), iv. 98.

OKLAHOMA.—Stillwater *Waugh*, no. 353 (an var. *humilis*?).

The following forms are doubtful:

WYOMING.—Fairbanks, *A. Nelson*, no. 337.

ARKANSAS.—Fulton, *Bush*, no. 2451.

ALABAMA.—Monte Sano, *Baker*, 23. v. 97 (glaberrima, foliis iis *A. glabrae* simillimus, sed calycis dentes ut in *A. fruticosa*).

FLORIDA (east).—Indian River, *Palmer*, no. 104 (et 1364) (foliolis ad 39!).

Var. *humilis*, var. nov.—*A. humilis* Tausch, Flora 21:750. 1838; the following names are also probably synonyms: *A. nonperforata*—Schkuhr, Bot. Handb. 2:333. 1808; *A. nana* Nutt. Fraser Catal. 1813, fide Torr. et Gray, et *A. nana* Sims, Bot. Mag. tab. 2112. 1820, haud Nutt. 1818; *A. fruticosa* Hayne, Dendrol. Flora 134. 1822, nec Linn.—Frutex *humilis*, foliis plerisque paullo minoribus saepe angustioribus iis *A. angustijoliae* haud dissimilibus.

As I said above, it is very difficult to decide whether a dried specimen belongs to typical *A. fruticosa* or to this variety or even to *A. angustijolia*, but the original specimens of TAUSCH are certainly not identical with *A. angustijolia*. The latter I never found living in our European gardens. Likewise the dwarf *humilis* everywhere appears within the limits of the type and perhaps only represents a mere "Standortsform."

Var. *crocealanata*, var. nov.—*A. crocealanata* Watson, Dendrol. Britann. 2: tab. 139. 1825; an *A. pumila* Tausch, Dendroth. Bohem. exsicc., nec Michx.?—Frutex elata omnibus partibus junioribus satis dense flavocanescensibus fere hirsutulis, foliolis etiam adultis superne sparsius subtus densim pubescentibus.

WATSON gives a very good description and drawing of his species, and the following forms agree very well with his characters:

MISSOURI.—Ocean Springs, *Miss Skeban* (?) 8. v. 95.

LOUISIANA.—Alexandria, *C. R. Ball*, no. 422.

ALABAMA.—Mobile, 16. iv (this specimen in the leaves somewhat resembles *A. herbacea*, but the calyx is quite that of *A. fruticosa*).

FLORIDA.—*Herb. Chapm.*, without other indication (this form has the leaflets in part somewhat more broadly elliptical); loco non indicato, *Herb. Chapm.* no. 1345.

Unfortunately all these specimens are in flower or with only very young fruits. It seems to me of great interest that such analogous forms appear in such very different localities.

12. A. *VIRGATA* Small, Bull. Torr. Bot. Club 21:17. 1894.—*A. georgiana* Small, MSS. in Herb.

GEORGIA.—DeKalb Co., Stone Mt., *Small*, 3 et 17. vii. 93; *Bilm. Herb.* no. 14c, 12. v. et 8. ix. 97; *Eggert*, 17. v. 99 (one of these forms with very large leaflets, 6.5×3.5cm, seems to me very near to *A. nitens* Boynt.) and 22. vii. 97; Cobb Co., *Harper*, no. 226.

NORTH CAROLINA.—Without locality or number, *Ashe* (distributed as *A. georgiana* Small).

FLORIDA.—Lake Co., Eustis, *Hitchcock*, vii. 94; *Nash*, no. 261 (see also the undetermined forms).

*A. virgata typica* is a species with rather broad ovate or elliptic-ovate leaflets; but there are the following southern forms (from Florida) which differ in smaller, more elliptic-lanceolate leaflets with more cuneate base. I cannot say yet whether they represent a good variety.

GEORGIA.—Gwinnett Co., *Small*, 20. vii. 93.

FLORIDA.—Near Jacksonville, *Curtiss*, no. 6410; Volusia Co., *Curtiss*, no. 6684; Clearwater, *Tracy*, no. 6870; Lemon City, *Tracy*, no. 7726; Lee Co., *Hitchcock*, no. 52.

In Biltm. Bot. Stud. 1:139. 1902, BOVNTON describes as a new species *A. nitens* from Waynesboro, Georgia. Unfortunately I have no type specimen at hand. It is possible that *A. nitens* is a good but very local species, distinguished by its glossy twigs, its thinner leaflets, and its more falcate nearly glandless legumes; but all these characters are rather slight ones.

I have before me two different specimens which I cannot identify with any other form mentioned here; the first probably may belong to *A. nitens*; the second is a very striking form, most resembling *A. paniculata* (*nervis foliolorum subtus elevatis*), but the leaflets are rather thin and glabrescent.

TENNESSEE.—Cocke Co., Kearney, no. 641 (also somewhat resembles *A. glabra*).

FLORIDA.—Chattahoochee River, *Bush*, no. 13.

### 13. *A. LAEVIGATA* Nutt., Torr. et Gray. Fl. N. Am. 1:306. 1838.

The type was found on the "banks of the Arkansas, near Salt River." The description given by NUTTALL is not clear. He says "leaflets distant, elliptical-oblong, attenuated below;" and also "with large distant and very obtuse leaflets." TORREY and GRAY did not see this species, and it seems to me very difficult to clear it up without an original specimen. The interpretation of *A. laevigata* given by BOVNTON in SMALL's *Flora* (625) is probably not correct; but if it is, *A. californica* and *A. laevigata* would be very nearly allied!

I myself believe that a specimen collected by an unknown gentleman on the banks of the Little Mamelle River, Arkansas, July 1825, which I found in Herb. Engelmann under no. 1043, with the determination *A. laevigata* Nutt., may be the true species of NUTTALL. Most of the leaflets are large and broad-elliptical with a round or slightly cuneate base. Some leaflets are more "suborbicularia" or more "late ovato-lanceolata." The apex is rounded and "fere semper leviter emarginata." The leaflets are rather thin, but firm and smooth and "fere glaberrima." The small fruits are the same as those of typical *A. texana*.

Another quite glabrous specimen is that which has been collected by E. Hall, no. 128, eastern Texas, Creek bank, Houston: foliolis minoribus, ad 3.5×1.8<sup>cm</sup> magnis, ellipticis, basis subrotundatis vel subcuneatis, apice rotundatis

vel emarginatis, mucronatis; fructibus maturis 5–6<sup>mm</sup> longis, dorso rectis, satis glanduloso-verrucosis.

Var. *PUBESCENS* Gray, Pl. Wright. 49. 1852.—*A. fruticosa* var. *subglabra* Gray, Jour. Bost. Soc. Nat. Hist. 6:174 (the first *subglabra* on this page!) 1850; *A. texana* Buckl. Proc. Philad. Acad. 452. 1861 (sed confer etiam Gray, eod. loco, p. 162. 1862!); *A. subglabra* Heller, Bot. Explor. South Texas 48. 1895.

The type of this variety is the same as *Lindheimer* specimen vi. 47, on a creek near Fredricksburg, Texas; according to my view this is only a variety of *A. laevigata*, if the latter is rightly interpreted above. GRAY (*l. c.*) and HELLER (*l. c.*) state that the true *A. texana* in no way differs from var. *pubescens* (=var. *subglabra* Gray, *l. c.*). I also believe that Heller's no. 1772 from Kerr Co., S. Texas, can be best taken as such a variety. There are some more pubescent forms which I name f. *mollis* (=*A. texana* var. *mollis* Boynt. Biltm. Bot. Stud. 1:139. 1902). To this form belongs a sterile specimen in Herb. Mo. Bot. Gard., with the printed statement "remnants of Buckley's Texas Herb.," and named in BOYNTON's hand "*laevigata pubescens* Gray."

14. *A. PANICULATA* Torr. et Gray, Fl. N. Am. 1:306. 1838.—*A. Roemeriana* Scheele, Linnaea 21:461. 1848.

TEXAS.—Lynchburg, *Lindheimer*, vii. 42 et (loco non indicato) no. 37 (anno 1843); Gladewater, *Reverchon*, no. 2665; Brazos, *Lindheimer*, no. 18 (anno 1843); Anderson Co., *Eggert*, 11. vi. 99; loco non indicato, *Pammel*, vii. 88; *Drummond*, no. 461 (vel. 261), anno 1835; Marshall, *Bush*, no. 991.

I conclude this account with the words of SCHLECHTENDAHL, which seem to me as true today as they were some fifty years ago: "Quae omnia fusius exposimus, ut inde pateat, quanta sit confusio in hoc genere et quam insufficiens status cognitionis nostrae."

VIENNA, AUSTRIA

## RUELLIA AND DIANTHERA: AN ANATOMICAL STUDY

THEO. HOLM

(WITH TWO PLATES AND THREE FIGURES)

Many genera of Acanthaceae have been studied from an anatomical point of view, and the literature is quite extensive. Much attention has been given, for instance, to the occurrence of the peculiar cystoliths, which seem to be especially characteristic of this family; the hairs, which are somewhat variable, have been studied also in a number of genera; and the several cases of anomalous stem-structure have attracted much attention. Suggestions as to an anatomical characterization of the family have been made by VESQUE,<sup>1</sup> but only in connection with the leaves. The anatomy of several Ruellieae has been described by OLGA TCHOUPROFF,<sup>2</sup> and quite a comprehensive anatomical monograph of *Thunbergia* has been presented by ROULET.<sup>3</sup> With the exception of these works, no others dealing with a general treatment of the family have appeared. Therefore, in spite of the rather copious literature, one does not find more than a few scattered notes on the stem structure in general, relatively very little about the leaf, and almost nothing about the roots. It must be remembered, however, that the authors, as a rule, had to depend on herbarium material or specimens cultivated in botanical gardens.

One species of *Ruellia* and one of *Dianthera* are very frequent in the vicinity of Washington, and since these species have not been examined so far, I studied them and found various points that may be of interest to students of plant anatomy. It will be seen from this study that *Ruellia* has a monostelic structure, which is typical of the stem of dicotyledons; while *Dianthera* has a polystelic axis. I was very much surprised in finding such structure in a member of

<sup>1</sup> Caractères des principales familles gamopétales tirés de l'anatomie de la feuille. Ann. Sci. Nat. Bot. VII. 1:326.

<sup>2</sup> Quelques notes sur l'anatomie systématique des Acanthacées. Bull. Herb. Boissier 3:550. 1895.

<sup>3</sup> Recherches sur l'anatomie comparée du genre *Thunbergia* L. fil. Bull. Herb. Boissier 2:259. 1894.

the Acanthaceae, since it is so very rare among dicotyledonous plants, being known only in *Nymphaea*, *Gunnera*, *Primula* (*AURICULA*), and *Pinguicula*. In polystelic axes, as described by VAN TIEGHEM and DOULIOT,<sup>4</sup> each stele has its own pith, parenchymatic rays, mestome, and endodermis, and they are all surrounded by a common cortical parenchyma. When the mestome (leptome and hadrome) is distributed in the axis as several isolated strands, and of mestome alone, such structure is called by the same authors astelic; a structure known in *Anemone*, *Ranunculus*, *Ficaria*, *Caltha*, *Hydrocleis*, etc.

In *Dianthera* the steles are very distinct and readily to be recognized as such, since they are cylindric and possess all the necessary elements. In comparing *Ruellia*, which is a land plant, with the aquatic *Dianthera*, they are found to be very different, and it is interesting to notice in the latter the combination of characters peculiar to aquatic plants in general and others peculiar to Acanthaceae.

#### *Ruellia ciliosa* Pursh.

A woodland type from open woods and thickets, associated with *Silphium trifoliatum*, *Salvia lyrata*, *Galactia pilosa*, etc. The stem above ground dies off in the late autumn (November) and the buds that winter over are subterranean. The rhizome, which persists for several years, is creeping and horizontal, or sometimes ascending; the horizontal direction I believe is most typical of the species, and in this the internodes are very short and almost concealed by the numerous thick roots. When the rhizome is ascending, the internodes are more or less stretched and readily visible. The leaves of the rhizome are very short, scale-like, and membranaceous. Several axillary buds are to be observed on the rhizome; in the larger of these, two or three internodes are frequently so distinct that the term shoots might be more correctly applied to these than buds; however, they remain underground during the winter.

The branching of the rhizome is sympodial, and very regularly so, even in old specimens. The apical internode terminates in an aerial shoot, which dies down to the ground at the close of the season, and the continued growth of the rhizome is secured by the develop-

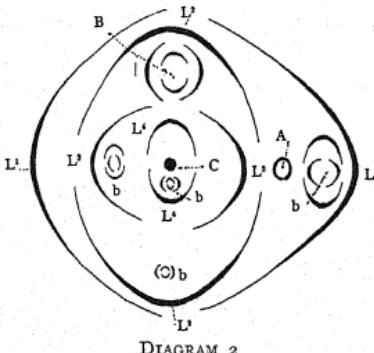
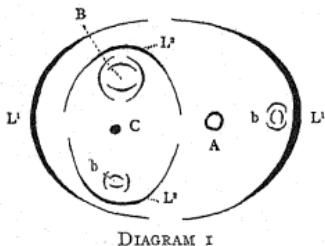
<sup>4</sup> Sur la polystélie. Ann. Sci. Nat. Bot. VII. 3:275.

ment of an axillary bud. In the accompanying diagram (*Diagram 1*), *A* represents the basal portion of the dead stem of 1905; *B*, the aerial shoot of 1906; *L<sup>1</sup>*, opposite leaves of the shoot *A*, and in the axil of the one of these (to the right) a small bud (*b*), while in the axil of the other the floral shoot (*C*) is developed. Shoot *C* has also a pair of

opposite, scale-like leaves (*L<sup>2</sup>*), both subtending buds, but of different size, the one behind *C* being the larger (*B*). This large bud (or young shoot) will grow into an aerial stem in 1907, while the two smaller buds (*b*) will remain dormant. The diagram is of a specimen in its fifth year, and not yet blooming.

If we examine the apical portion of a mature specimen with fruits (early in October), the position of the buds is the same as described above, but the number of leaves may be larger. In *Diagram 2* the dead stem *A* has one pair of opposite leaves (*L<sup>1</sup>*), the one to the right subtending a bud (*b*), while the one to the left subtends a floral shoot (*C*). This floral shoot bears three pairs of leaves with one large (*B*) and three smaller buds (*b*); *B* corresponds with *B* in the diagram described above, and is situated behind *C*. But *B* is here separated from *C* by two pairs of leaves, and thus is situated somewhat lower than in the other specimen,

but occupies the same position behind the floral shoot *C*. The large bud in the axil of *L<sup>1</sup>* (to the right of *A*), as stated in the preceding, is so far advanced that the internodes are quite distinct, and this bud sometimes develops into an aerial shoot when *C* becomes injured. Otherwise the rhizome does not branch so as to develop



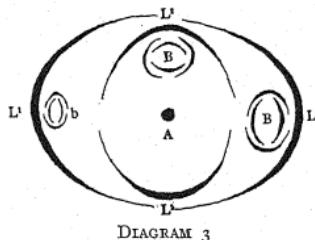
more than one aerial shoot at the same time; some few specimens I have found, however, where two flowering stems were developed, one of these apical, the other from a bud of a much older portion of the rhizome. Two to five roots develop from the internodes or directly beneath the buds at the nodes.

The rhizome of *Ruellia* is thus relatively short, when it grows in a horizontal direction, and its power to wander is very limited. Nevertheless one may find several specimens with vegetative shoots in connection with a single rhizome and at some distance from it; such cases are not very rare, but these shoots do not arise from the rhizome, but from roots, and are thus simply root-shoots. The diagram of a root-shoot in its first year (*diagram 3*) shows two pairs of scale-like leaves, one central vegetative shoot (*A*), one large bud (*B*) behind this, and another one (*B*) to the right, while a smaller one (*b*) is situated in the axil of *L<sup>1</sup>* to the left of *A*. The position of the buds is essentially the same as in the other diagrams, with the exception that the aerial shoot (*A*) occupies here the center of the diagram and the apex of the young rhizome, since this is in its first year of growth.

It is readily seen from these diagrams that in young specimens, in this case a root-shoot, the aerial stem terminates the rhizome in the first year of its growth (*diagram 3*). In older rhizomes, on the other hand, the aerial stem terminates only the apical internode and has actually arisen from the axil of a leaf borne upon the basal internode of the shoot of the preceding year. The shoot *A* is thus always terminal, and always the mother shoot of *C*.

The occurrence of root-shoots has not been recorded for Acanthaceae; at least they are not included in WITTRICK's extensive list of "plants with root-shoots."<sup>5</sup> Three types of such shoots are proposed by WITTRICK: reparative, additional, and necessary; those of

<sup>5</sup> Om rotstökt hos örtartade växter. Förhdigr. Botan. Sällsk. Stockholm. 1883. [Bot. Notiser 1884:1.]



Ruellia belong to the second type, the additional, which develop spontaneously upon the uninjured plant, and which constitute an important addition to the subterranean vegetative organs. This category of root-shoots is the one that is best known and most frequent; our species of *Apocynum* illustrate this manner of propagation in a striking degree, and I have seen as many as ten flowering stems of *A. cannabinum* developed upon one single root;<sup>6</sup> it is also the kind so commonly met with in Sassafras, *Rhus Toxicodendron*, Comandra, etc. It may be stated at the same time that the development of additional root-shoots appears to be characteristic of plants in certain localities; for instance, on the plains of Colorado I noticed a remarkably large percentage of plants of various families that exhibited this peculiar mode of propagation.

#### THE ROOTS

In mature specimens the secondary roots are quite thick, somewhat fleshy, and of a light brown color. The epidermis is thin-walled and somewhat hairy; it covers an exodermis of one layer, which is also thin-walled and whose cells show the same lumen and shape as those of the epidermis. The cortex is differentiated into three distinct zones: a peripheral of three layers of more or less thick-walled cells with many cystoliths; about nine strata inside the peripheral, which consist of thin-walled cells with rhombic intercellular spaces, and of very thick-walled sclerotic cells (figs. 1, 2) besides cystoliths, while no raphids were seen, and no deposits of starch; the innermost stratum, represented by a thin-walled endodermis of small cells and with the Casparyan spots very plainly visible.

The sclerotic cells are very characteristic and readily noticeable by their narrow lumen and porous walls; they are stained bright yellow by chlor-zinc-iodin. Similar sclerotic cells are known also from the roots of *Thunbergia annua*, according to ROULET (p. 343). The cystoliths of the Acanthaceae are very well known, and have been described very carefully by SOLEREDEER (p. 697). They vary somewhat in outline, from almost quadrangular to fusiform with the

<sup>6</sup> That the horizontal shoot-bearing roots of *Apocynum* have been mistaken for "horizontal rootstocks" may be seen in MILLER: Dogbanes of District of Columbia, Proc. Biol. Soc. Washington 13:79.

ends obtuse or acute; their surface, however, is always very distinctly granular. Viewed in longitudinal sections the cells containing cystoliths are usually shorter than the sclerotic. The cortical parenchyma of Ruellia thus shows a very characteristic structure.<sup>7</sup>

Inside the endodermis is a thin-walled, continuous pericambium, which surrounds the mestome with its mechanical support of stereome. There are four strands of stereome, each consisting of five to twelve typical stereome cells in one or two layers between the pericambium and the leptome. The leptome itself shows the presence of raphidines, but they seem to be very scarce in this species of Ruellia, since I never found more than one in each cell, and only in a very few places. Raphidines were first described by Russow,<sup>8</sup> who observed them in the root, stem, and leaf of *Hexacentris coccinea*, etc. VESQUE<sup>9</sup> has also described them as characteristic of Cyrtanthera, Meninia, Adhatoda, and Fittonia; while ROULET (*l. c.*) found them in a number of species of Thunbergia. They are also to be observed in Mendoncia and Pseudocalyx, according to RADLKOFER.

The hadrome contains numerous vessels which extend to the center of the root, but secondary formations had already commenced so that the primitive organization of the stele could not be ascertained; however, the four strands of stereome may indicate a tetrarchic structure. Although the specimens of Ruellia were collected as late as the third week of September, no deposits of starch were observed in the roots.

Besides the thick, somewhat fleshy secondary roots, there are some that are quite thin on account of the less development of cortex, which consists only of three strata, thin-walled throughout, with no

<sup>7</sup> A cortex of exactly the same structure has been described and figured by HENRY G. GREENISH (Pharm. Jour. and Trans. London 1891:839) as representing that of the root of what he took to be *Phlox carolina*. However, the root of that species does not possess cystoliths or sclerotic cells, nor do the roots of any other species of Phlox that I have examined. The rhizomes and roots which GREENISH had before him, and which were sold as those of *Spigelia marilandica*, no doubt belonged to our Ruellia, a plant that has about the same distribution as Spigelia, and whose rhizomes somewhat resemble those of Spigelia. Hence the statement that cystoliths occur in the root of *Phlox carolina* (SOLEREDER, p. 622) may be safely omitted.

<sup>8</sup> Sitzungsber. Naturf.-Gesells. Dorpat 5:308. 1878-1880.

<sup>9</sup> Sur quelques formations cellulosiennes locales. Ann. Sci. Nat. Bot. VI. II: 181.

sclerotic cells, but with a few cystoliths. The stele is composed of three rays of hadrome, and no secondary growth was yet observable.

The capillary, lateral roots are more hairy than the mother root; they have an exodermis whose cells are larger than those of the epidermis and of the peripheral stratum of cortex. The cortical parenchyma is very thin-walled and contains neither cystoliths nor sclerotic cells; the stele shows two rays of vessels with thin-walled conjunctive tissue; but no stereids and no raphidines were observed in these very thin roots.

The roots of the Acanthaceae have been studied only from a relatively small number of genera, and it seems very likely that the structure described above may be observed in several other species of Ruellia and in many other genera of the family, as already indicated in the works of RADLKOFER, ROULET, RUSSOW, and VESQUE.

#### THE RHIZOME

As stated above, internodes of the rhizome are either very short and horizontal, or stretched and ascending. The latter form shows the following structure: the epidermis is perfectly glabrous and smooth, with the outer cell walls thickened, and some of the cells containing cystoliths; three continuous layers of collenchyma with cystoliths surround the cortex, which consists of five strata, whose thin-walled cells increase in size toward endodermis and contain many cystoliths and raphids, but only a very few sclerotic cells, and no starch; the endodermis is thin-walled and the cells are much smaller than those of the adjoining cortical parenchyma; a few stereids are located inside the endodermis; and the stele shows an almost confluent zone of leptome and numerous rays of vessels (about ten in each row) with narrow parenchymatic rays; a broad, thin-walled pith occupies the central portion of the stele, in which cystoliths and deposits of starch were observed. A longitudinal section of the internode shows the raphid cells to be very narrow, but much longer than the cells of the cortex proper; the cystoliths vary in length, those of the collenchyma being longer than those of the cortical parenchyma, and obtuse or attenuate at each end.

## THE STEM ABOVE GROUND

The basal internodes are obtusely quadrangular and hairy with glandular or pointed hairs of the same structure as those of the leaves (*figs. 3, 4*). The cuticle is smooth, and the cells of epidermis are small, somewhat thickened on the outer wall, and contain cystoliths. Five layers of thick-walled collenchyma surround the cortex, which is thin-walled and consists of about nine strata with wide rhombic intercellular spaces; cystoliths and raphids occur in the cortex, but no sclerotic cells and no deposits of starch. A thin-walled endodermis surrounds the stele, which is obtusely quadrangular in cross-section. A few scattered stereomatic cells are located inside the endodermis and outside the leptome. The mestome strands are collateral; the hadrome is represented by short rays of vessels, separated by mostly four rows of thick-walled parenchyma; a few layers of cambium are observable between the leptome and hadrome, but none outside the medullary rays. The central portion of the stele is occupied by a thin-walled pith of large cells, several of which contain raphids and crystals of various forms, prismatic, rhombic, etc., but no cystoliths and no starch.

The structure of the apical internodes differs in a few points from that of the basal portion of the stem. For instance, the collenchyma occurs as separate strands interrupted by the cortex, which is rich in chlorophyll but contains no cystoliths. Moreover, the stereome is better developed in the superior internodes, forming an almost closed sheath inside the endodermis, but outside the collateral mestome bundles.

In the nodes there is a much stronger development of the collenchyma than in the internodes, and the cortex does not break through this tissue so as to reach the epidermis. By comparing the structure of a node with that of the internode below, the following deviations were noticed: there are ten continuous layers of collenchyma in the node, but only five and not continuous in the internode; there are seven layers of cortex in the node, but only five in the internode. The stereome inside the endodermis is reduced to a very few cells in the node, while it is much better represented in the internode. The pith occupies a larger portion of the section in the node than in the internodes. Finally, the cells of the cortex and pith show a wider

lumen in the node than in the internode. The nodes of other genera of the same family have been described by RÜTZOU,<sup>10</sup> who found raphidines in the leptome of *Schaueria* and *Beloperone*.

#### THE LEAF

The ample blade is hairy on both faces, especially on the lower, and along the margins; the ventral face is very smooth, while the dorsal shows a prominent keel from the midrib, and also some of the secondaries (the basal) project on this face. The structure is bifacial in respect to the distribution of stomata and the differentiation of the chlorenchyma.

A very simple and uniform structure is to be observed in the cuticle, which is quite thin and smooth on all parts of the blade; and with the exception of the occurrence of the stomata only on the lower face, the structure of the epidermis is identical on both faces. The lateral cell walls are prominently undulate (*fig. 3*) in the ordinary epidermal cells, or straight in those that contain cystoliths (*fig. 3*). Viewed *en face*, the cells containing cystoliths are readily distinguished by their long and narrow outline and by their contents; they abound especially on the dorsal face, underneath the collenchyma, but they are also very frequent between the veins, above and below the chlorenchyma. Glandular hairs with round pluricellular heads are scattered on both faces, but not so frequent as the very long pointed ones (*fig. 4*). The stomata have two subsidiary cells vertical on the stoma, a structure that is also characteristic of certain genera of Caryophyllaceae. Viewed in transverse sections, the cells of the epidermis are quite large, especially above the palisade tissue; but a considerable decrease in lumen is noticeable in that part of the epidermis that covers the collenchyma, that is above and below the larger veins, where the outer cell wall also becomes moderately thickened. The stomata are raised a little above the adjoining epidermis, and the air-chamber is deep and wide.

The mechanical tissue is represented only by hypodermal strands of a few layers of thick-walled collenchyma, which follow the stronger veins, especially the midrib. No stereome was observed. A thin-walled water-storage tissue occupies the greater portion of the promi-

<sup>10</sup> Om Axeknuder. Bot. Tidsskr. Kjbhvn. 1880-1: 257.

nent midrib, above and below the mestome bundles. This tissue is also represented, though less developed, on the leptome side of the secondary veins, but only the stronger ones that proceed from the base of the midrib.

In proportion to the size of the leaf-blade, the chlorenchyma is very thin and consists of one layer of typical palisades on the ventral face, and of an open pneumatic tissue on the dorsal; the latter is composed of two to three strata of oblong or more or less roundish cells with very wide intercellular spaces. The chlorenchyma is partly interrupted by the collenchyma above the midrib, but the palisades extend for some distance through the water-storage tissue above the mestome bundles of the midrib (fig. 6). According to VESQUE (*l. c.*, p. 333), the palisade tissue is continuous, that is, not interrupted by collenchyma, in *Ruellia maculata* and *R. formosa*, in various species of *Eranthemum*, *Strobilanthes*, *Beloperone*, etc., but interrupted, as described above, in *Sanchezia*, *Ebermayera*, etc.

All the mestome bundles are collateral; those that constitute the midrib are arranged in a broad arch with the concave face turning upward; the others follow the chlorenchyma in one plane. Only the smallest ones, which are not accompanied by water-storage tissue, have a closed colorless and thin-walled parenchyma sheath. The midrib is composed of several (mostly five) mestome strands, separated from each other by strata pertaining to the water-storage tissue; the hadrome consists of a few rows of rather narrow vessels, with three to four in each row. The leptome (fig. 5) is well-represented as several groups underneath the hadrome, and also occurs as small, isolated strands between the hadromatic rays (*L* in fig. 5). No closed parenchyma sheath is observable in the midrib, but there is nevertheless an endodermis. This very characteristic sheath is represented here on the leptome side only; it is thin-walled, like the adjoining water-storage tissue, and shows very plainly the spots named after Caspary. Without forming a closed sheath, the endodermis thus forms an arch parallel with the keel, and ceases outside the leptome of the two outermost mestome strands. A like structure in regard to the open endodermis in the midrib is described by ROULET (*l. c.*) as characteristic of certain species of *Thunbergia*, for instance, *T. fragrans*, *T. hastata*, and *T. hirta*.

The portion of the blade described above was near the middle, where the leaf is broadest; however, the structure is essentially the same throughout. At the narrow apex there is only a decrease in width of the collenchyma, the water-storage tissue, and the midrib, which consists only of three almost confluent mestome strands; while the chlorenchyma has not undergone any change.

Characteristic of the leaf of our *Ruellia*, therefore, is the interruption of the chlorenchyma by the collenchyma, and the presence of isolated leptomatic strands between the rays of hadrome. The structure of the stomata, the presence of glandular and long pointed hairs, and the cystoliths are, so far as we know, characteristic of Acanthaceae in general.

#### THE PETIOLE

A transverse section of the very short petiole shows an approximately crescent-shaped outline, the ventral face being almost flat in contrast with the dorsal, which is obtusely carinate. The structure is almost identical with that of the midrib in the blade. Glandular and pluricellular pointed hairs occur also, and two or three strata of thick-walled collenchyma follow the outline inside the epidermis. A colorless, thin-walled water-storage tissue occupies the greater portion of the section, and the mestome bundles are arranged in an arch as in the midrib, while two very small strands of mestome are located in the thin margins, surrounded by chlorophyll-bearing parenchyma.

Cystoliths are abundant in the epidermis, but are absent from the colorless parenchyma, in which only raphids and some long prismatic crystals were noticed. On one side of the broad arch-shaped midrib two wide sclerotic cells were observed, and a few raphidines in the leptome.

#### *Dianthera americana* L.

This species is very frequent in the vicinity of Washington, D. C., where it grows in creeks and in the Potomac River, generally associated with *Saururus*, *Sagittaria*, *Pontederia cordata*, etc. It has a very long, horizontally creeping rhizome, with opposite, scale-like leaves and stretched internodes of an obtusely hexagonal outline. Secondary roots, from four to eight or even more, are developed at

the nodes, but not directly underneath the leaves, mostly between them; the roots are quite thick and long, and branch freely. The rhizome represents a sympodium, being terminated by a floral shoot, which dies off at the end of the first season; axillary buds occur on the rhizome and at the base of the aerial shoot, and these winter over and continue the growth of the rhizome at the beginning of the next season.

The germination seems to be unknown, and I regret to say that I have never succeeded in studying the young stages of this common plant.

#### THE ROOTS

As described above, the rhizome is amply provided with roots (secondary), that are quite thick and much branched; they remain active for at least two years and represent a combination of storage and nutritive roots, but are not contractile. The internal structure of these secondary roots exhibits very plainly the features characteristic of roots of aquatic plants. The epidermis is very small-celled and glabrous. The exodermis consists of three very distinct strata (fig. 8) of polygonal cells with the walls slightly thickened and deep brown, but with no foldings. The cortical parenchyma is thin-walled and consists of about seventeen layers, the cells very regularly radially arranged and traversed by wide lacunes from the radial collapsing of several of the strata; the outermost layer of the cortex and the innermost three usually remain intact. Small quantities of starch, but no crystals, were observed in the cortex. The stele shows a thin-walled endodermis and pericambium, which was never found to be interrupted by the proto-hadrome vessels. Although these roots have increased in thickness, the primitive organization is yet quite distinct, there being mostly five rays of hadrome with old vessels. The cambial arches inside the leptome (fig. 7) are readily visible, as well as the very thin-walled young vessels. The leptome is well represented, and in several cases it appeared as if there were two proto-leptome cells in each strand. A thin-walled conjunctive tissue occupies the inner portion of the stele like a central pith.

No crystals were observed in any parts of these roots, nor any sclerotic or stereomatic cells; in the leptome, however, a few raphidines were noticed, but only in some of the roots. By studying the

same root at different places, I noticed that the collapsing of the cortex does not take place in the entire length of the root, but only in the basal (thickest) portion. The occurrence of rhipidines is very variable; in some roots they were observed only in the younger apical portion; in others they were found also in the basal.

The lateral roots of the first order show the same structure as the mother root, but the number of strata in exodermis and cortex is generally smaller. The exodermis, for instance, may be represented by only a single layer, but sometimes three are found, as in the mother root. The cortex may be solid or collapsed, and may be reduced to only three strata. The epidermis does not differ from that of the secondary root, and is glabrous. The endodermis is thin-walled, and the secondary formations are usually not so advanced but that the primitive organization may be readily observed to be diarchic or tetrarchic.

A corresponding structure is to be found in the lateral roots of the second order. They are capillary and have a glabrous epidermis and an exodermis of two layers. The cortex consists of three thin-walled strata, which are collapsed at the base of the root, but entire near the apex. Inside the continuous pericambium are a few stereometric cells, mostly only two, diametrically opposite each other. The leptome is represented by two broad strands alternating with two rays of hadrome with two wide reticulate vessels in the center, and about three scalariform outside. No secondary growth takes place in these roots.

If it were not for the presence of raphidines, the root of *Dianthera* would be utterly unlike that of *Ruellia* described above. Moreover, the habitat of *Dianthera* is naturally the cause of the very different development of exodermis and cortex, and of the absence of cystoliths, sclerotic cells, and crystals.

#### THE STEM

The rhizome with the stolons and the ascending aerial shoots show the same structure in general, and, as stated above, the axis is polystelic.<sup>11</sup> This peculiar structure was observed in all the inter-

<sup>11</sup> A polystelic stem is also possessed by *Dianthera crassifolia* Chapm. and *D. lanceolata* (Chapm.) Small, but not by the following species: *D. comata* L. (Porto

nodes of the rhizome and of the ascending shoots, in the slender as well as in the more or less swollen portions of the vegetative axis. In the nodes the polystelic structure becomes somewhat obscured by the anastomoses of the steles; nevertheless, the presence of several steles is usually recognizable.

In outline the rhizome is obtusely hexagonal and smooth; no stomata, but small, sessile glandular hairs were observed. Formations of cork develop from the epidermis, including the cells with cystoliths (fig. 16). Inside the epidermis there is a continuous zone of very thick-walled collenchyma of about six layers, bordering on the cortical parenchyma. The cortex consists of numerous strata of thin-walled cells, which are round in transverse sections and with relatively wide intercellular spaces (fig. 17); starch was observed in the cortex, and numerous bundles of true raphids, but no chlorophyll. Throughout the internodes of the rhizome (and also of the aerial shoots) the cortical parenchyma represents a homogeneous tissue from the periphery to the central portion, whether a central stele is present or not. No central pith is differentiated in the internodes except where there is a central stele, also a small tissue of this parenchyma is to be found in the peripheral steles. It appears as if the horizontal internodes of the rhizome have always six peripheral and one central stele; these steles are cylindrical and each is surrounded by a thin-walled endodermis with the Casparyan spots plainly visible and with deposits of starch. Inside the endodermis are a few cells of thick-walled stereome, but too few to form a sheath. In the peripheral steles the leptome and hadrome form an almost complete ring, with a small central pith; in the central stele, on the other hand, the mestome is arranged in two arches, with a broad parenchyma in the middle.

Between the leptome and hadrome are a few strata of cambium, and between the strands of mestome are three or four rays of thick-walled parenchyma. Raphidines (fig. 18) were observed very frequently in the leptome.

Rico), *D. glabra* B. et H. (Costa Rica), *D. incerta* Brandg. (Lower Calif.), *D. ovata* Walt. (Florida), *D. parvifolia* B. et H. (Texas), *D. pectoralis* Murr. (St. Croix), and *D. sessilis* Gray (St. Domingo). Of these nine species I had only dried material, which was quite sufficient for a study of the steles, but not of the anatomical structure in general.

In a stolon, terminated by an over-wintering bud, I found the same structure as described above as characteristic of the main rhizome, the only difference being that the collenchyma was less thick-walled; that the cortex contained large deposits of starch; and that the central stele showed very prominently three arches of mestome (leptome, cambium, and hadrome) separated from each other by very broad rays of thin-walled parenchyma. While the hadrome in this stele was thus confined to three arches of collateral mestome strands, the leptome was developed as isolated groups inside the endodermis, not only outside the hadromatic rays, but also between them.

While thus the peripheral steles of the main rhizome and the stolons show an almost equal development of collateral mestome strands around a central pith, the stele in the center shows the hadrome more or less distinctly combined in arches (two or three), while the leptome follows the entire periphery inside the endodermis (*fig. 14*).

A corresponding structure is to be found in the aerial shoot which bears the long, narrow leaves and the inflorescences. In the apical internodes, for instance, we find the same tissues and arranged in the same manner, but the change of medium naturally brings about certain modifications of structure. In the rhizome and in the submerged portion of the ascending shoots the cortex does not reach the epidermis, but is completely inclosed by the collenchyma. In the aerial internodes, on the other hand, the cortex extends to the epidermis, thus breaking through the collenchyma, which here becomes developed as isolated strands, as prominent ridges; where the cortex reaches the epidermis it develops typical palisades, which contain much chlorophyll (*fig. 21, C*).

The cuticle is very smooth and thin in the aerial internodes. The epidermis has stomata and glandular hairs, and contains cystoliths. The collenchyma is very thick-walled and forms eight prominent strands of about ten layers; six of these strands correspond with the six peripheral steles, while the remaining two, which are somewhat smaller than the others, are diametrically opposite each other and between two steles. The cortex is differentiated into one or two strata of palisades, located in the furrows between the collenchymatic ridges; the inner portion of the cortex contains less chlorophyll

and the cells are more roundish in cross-sections; while the innermost, which occupies the greater portion of the internode, is a very open tissue with no chlorophyll, but with some small deposits of starch. The intercellular spaces are quite wide and the general structure of the cortex agrees with that of an aquatic plant. Six peripheral steles are located inside the six collenchymatic strands, while a seventh occupies the center of the internode. They are all orbicular in transverse sections and each has a thin-walled, completely closed endodermis, inside of which is another sheath of about two layers of thin-walled stereomatic tissue. The mestome bundles of the peripheral steles are collateral and arranged in an arch toward the periphery of the stem, while the inner face of the steles is occupied by a pith and a few scattered strands of pure leptome (*fig. 14, L*). The central stele shows the same structure as the peripheral. *Fig. 13* shows the mestome of one of the peripheral steles; there are a few strata of cambium inside the leptome, and some young (three) vessels besides older ones in two rays with thin-walled parenchyma between. The node above this internode shows at once a change in the disposition of the steles, that is, four of the peripheral have fused together in pairs so as to form two large steles of triangular outline, while the central and the remaining two peripheral are unchanged. The steles that thus fuse together in the node are those of the internode which are marked *S* in *fig. 9*, and between which there is an isolated strand of collenchyma.

In this node the thin-walled collenchyma surrounds the cortex completely, which is here almost destitute of chlorophyll and which represents a more compact tissue than in the internode below. No raphidines were observed in the node, but many crystals of various kinds, needle-shaped, rhombic, prismatic, etc., abound in the pith of the broader steles.

A dilation of some of the steles thus takes place in the node, resulting in the gradual concrescence of two steles on each side of the node. From the union of these steles each of the two opposite leaves receives three mestome cylinders, readily observed in the petiole as one central, very broad, and arch-shaped cylindré, with a much smaller one on either side. By following the structure of several internodes of a single shoot, I noticed that the central stele is not

present in all of them. Sometimes this central stele was observed in every other internode, but I found also some cases where the apical and basal internodes possessed such a central stele, but not the two internodes between them.

In the basal internodes of the aerial shoots the structure of the stele becomes more regularly developed, the leptome and hadrome forming an equally broad ring around the central pith, which consequently becomes much reduced in width; in other words, these steles show exactly the same concentric and uniform structure as the central stele of the monostelic axes.

In very small vegetative shoots, developed in the axils of the leaves of the larger floral ones, the structure is as described above, but the number of steles may vary. For instance, in a shoot consisting only of two short internodes, the basal one contained only four steles, all peripheral, while the upper one had one central, two large peripheral, and two still broader ones, each of which consisted of two that had partly fused together; the latter corresponding in position with the four marked *S* in *fig. 9*. In these internodes the structure and disposition of the collenchyma and cortex was exactly as described above.

In order to ascertain the number of steles in the stem of *Dianthera*, I examined shoots of very different height and thickness, but the number never exceeded seven—six peripheral and one central. A very thick branch with two opposite and equally robust shoots showed seven steles above and below the node, and also in the two lateral shoots. It is somewhat surprising to see that this polystelic structure ceases at once when the axillary shoot is an inflorescence and not a vegetative branch. The short inflorescence of *Dianthera americana* is borne upon a long, naked internode, and this internode is monostelic from base to apex. In this the cortex incloses completely one broad central cylinder with one zone of leptome and one of hadrome, with parenchymatic rays of equal size and with a central cylindric pith, as is characteristic of monostelic axes.

In the genus *Primula* the Danish botanist VAUPELL<sup>12</sup> detected the polystelic structure in *P. Auricula*, and VAN TIEGHEM and DOULIOT

<sup>12</sup> Om Rhizomets Bygning hos *Primula auricula* og *chinensis*. Vidensk. Medd. Naturhist. For. Kjöbenhavn 1849:76; and Untersuchungen über das peripherische Wachsthum der Gefäßbündel der dicotylenen Rhizome. Leipzig. 1855.

(*l. c.*) have extended his observation to a large number of species of various sections of the genus. One of the results of their investigation is that a complete accordance exists between the external and internal structure of these species of *Primula*. These authors proposed the reestablishment of TOURNEFORT's genera *Primula* and *Auricula*, the former characterized by possessing a monostelic, the latter a polystelic, axis. Whether this same peculiarity in the stem structure, which I have found in *Dianthera americana*, *D. crassifolia*, and *D. lanceolata*, may be used in classifying the species under two distinct genera, I must leave to future investigators to decide, who have access to more abundant material.

#### THE LEAF

The narrowly lanceolate and almost sessile leaves are held in a vertical position above the water; they are smooth and glabrous on both faces, while along the margins unicellular, short, pointed hairs occur; over these hairs the cuticle is granulous, but smooth and thin above the epidermis of both faces of the blade. There are only a very few of these hairs, and they were observed only on young leaves.

The leaf is neither exactly dorsiventral nor unilateral, when the distribution of stomata and the structure of the chlorenchyma are considered; the stomata are just as frequent on the ventral as on the dorsal face, and some palisade cells occur here and there inside the dorsal epidermis. Viewed *en face* the cuticle shows no striations, and the lateral cell walls of the epidermis are somewhat undulate on the dorsal face, but almost straight on the ventral; small glandular hairs (*fig. 10*) occur on both faces of the blade. The stomata are not arranged in any way so as to be parallel with the longitudinal axis of the leaf; and they have two subsidiary cells vertical on the stoma. *Fig. 12* shows the commonest form of the outline of the subsidiary cells, which seems to be less regular and more variable on the upper face of the leaf than on the lower. Viewed in transverse section, the outer wall of the epidermis is slightly thickened, and the lumen of the cells is wider on the upper face; the stomata are raised a little above the adjoining epidermis, and have a deep and wide air-chamber. In the lateral parts of the blade, which are much thinner than the median with its broad and thick midrib, the chlorenchyma (*fig. 22*)

shows one layer of typical palisades above a pneumatic tissue of roundish to more or less oblong cells with wide intercellular spaces. However, the stratum of pneumatic tissue which borders on the dorsal epidermis is in some places developed almost as palisades, the cells being more regularly oblong and more compact; as will be shown later, a palisade tissue is very typically represented on both sides of the dorsal keel of the midrib. No crystals were observed in the mesophyll, but long fusiform cystoliths are frequent in the epidermis, especially on the dorsal face.

Collenchyma and stereome seem to be entirely absent from the lateral portion of the blade, and thus the lateral veins are directly imbedded in the chlorenchyma. They are very fine, orbicular in cross-section, and surrounded by a thin-walled, colorless parenchyma sheath.

A more complicated structure is to be found in the median portion of the blade with the strong midrib. Near this part of the blade the leaf is thicker, due to an increase in height of the palisade tissue, while in the midrib a prominent broad and very obtuse keel is to be observed on the dorsal face, and a similar, though much less projecting one, on the ventral. The epidermis is very small-celled in this part of the blade and covers a broad strand of thick-walled collenchyma in about three layers on the dorsal face. There is also collenchyma on the ventral face, and this is extremely thick-walled and represented either by two separate strands near the middle, or by a single one exactly above the center of the midrib. A large, thin-walled colorless tissue occupies the greater portion of the keel, but occurs also above the mestome bundles which compose the midrib. The chlorenchyma is on faces of the midrib interrupted by the collenchyma; it occurs as two strata of high palisades on the ventral face, and as one or two strata of lower palisades (*fig. 23*) in the sides of the keel; thus, as stated above, this tissue is not confined to the ventral part of the blade alone. The pneumatic tissue is also well developed, but only near the dorsal keel. There are three mestome bundles in the midrib; one very broad, in transverse section crescent-shaped with the concave face turned upward, and two very small lateral ones, orbicular in cross-section. A thin-walled endodermis covers the leptome side of these three mestome strands, but not the hadrome.

In the middle there is an arch of thin-walled stereome in two or three layers between the endodermis and the leptome. The leptome has very narrow sieve tubes and companion cells, and the hadrome consists of rather narrow vessels in many short rays. In the two small mestome bundles the mestome is less developed, and there is no stereome. Raphidines abound in the leptome of all three veins, sometimes filling the cells completely.

There are thus in the leaf of *Dianthera* several features indicating the immediate relationship of the genus, illustrated by the structure of the stomata and the presence of raphidines and cystoliths. The distribution of stomata and palisades on both faces of the leaf at least points toward the general structure of an aquatic plant.

#### THE PETIOLE

There is a short but very distinct petiole, which is approximately hemi-cylindric in outline, the convex face being the dorsal. The internal structure is almost identical with that of the midrib. There are the same tissues of the same development, but distributed in a somewhat different way; for instance, the collenchyma surrounds the colorless parenchyma almost completely, being interrupted only by one narrow group of palisades in the middle of the ventral face, and by two small groups of the same tissue on the sides of the carinate dorsal, and very near its edges. There are three mestome strands in the center, the outline and minor structure of which agree in all respects with the midrib. The petiole has no stereome; cystoliths were found in the epidermis and raphidines in the leptome.

#### Summary

With the exception of the polystelic axis in *Dianthera*, the structure of the two plants agrees well with that of Acanthaceae in general as recorded by SOLEREDER, namely, the type of stomata, the glandular and simple hairs, the cystoliths, the raphidines, the various crystals of calcium oxalate, etc. These characters are some of the most important for distinguishing the family.

Although *Ruellia* and *Dianthera* possess the principal characteristics of the family, the very different environment in which they live has been the immediate cause of several modifications in structure.

In *Ruellia* the structure is that of a land plant as follows: the hairy root with a solid cortex and with many sclerotic cells; the very hairy stem with a closed sheath of stereome inside the endodermis; the very hairy leaves held in a horizontal position and with a strictly bifacial structure in regard to stomata and palisade tissue. In the aquatic *Dianthera*, on the other hand, the roots are glabrous (at least in adult specimens) and the cortex is collapsed; the stem has only small, glandular hairs and the cortex shows intercellular spaces of sometimes enormous width; the narrow leaves are held in a vertical position and are approximately isolateral, so far as concerns the stomata and palisade cells; in addition to the glandular hairs only very short and mostly unicellular hairs cover the margins of the younger leaves.

BROOKLAND, D. C.

#### EXPLANATION OF PLATES XI AND XII

##### *Ruellia ciliosa*

###### PLATE XI

FIG. 1. Transverse section of root, showing sclerotic cells in the cortical parenchyma.  $\times 320$ .

FIG. 2. Longitudinal section of a sclerotic cell.  $\times 320$ .

FIG. 3. Epidermis of lower face of leaf, showing two stomata, a cystolith, and a glandular hair, viewed *en face*.  $\times 320$ .

FIG. 4. Hair from lower face of leaf.  $\times 240$ .

FIG. 5. Transverse section of part of midrib of leaf, showing the leptome extending between the hadromat rays: *end*, endodermis; *l*, leptome; *h*, hadrome.  $\times 560$ .

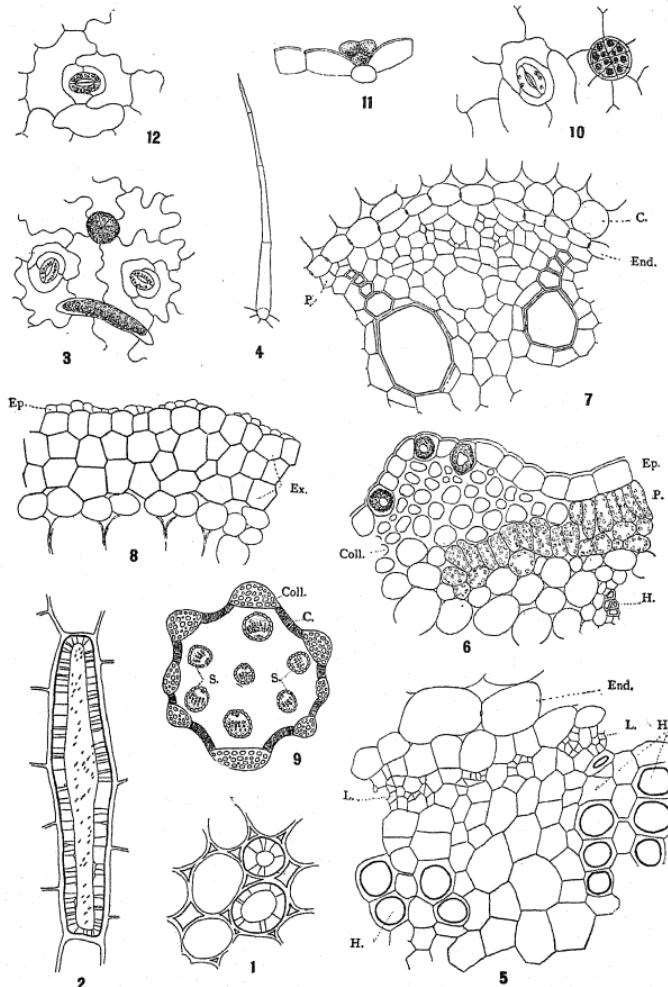
FIG. 6. Transverse section of part of leaf, showing the palisade tissue (*p*) extending above the midrib: *h*, the hadrome of the small lateral mestome bundle; *coll*, collenchyma.  $\times 230$ .

##### *Dianthera americana*

FIG. 7. Transverse section of a secondary root, showing the beginning of growth in thickness: *c*, cortex; *end*, endodermis; *p*, pericambium; between the two rays of vessels and inside the leptome is an arch of cambial tissue and a young, wide, and very thin-walled vessel.  $\times 496$ .

FIG. 8. Transverse section of a secondary root: *ep*, epidermis; *ex*, exodermis of three layers.  $\times 320$ .

FIG. 9. Transverse section of stem internode, showing eight strands of collenchyma (*coll*) separated by palisades, the peripheral strata of cortex (*c*), and



*Auctor ad nat. detin.*

HOLM on RUELLIA and DIANTHERA

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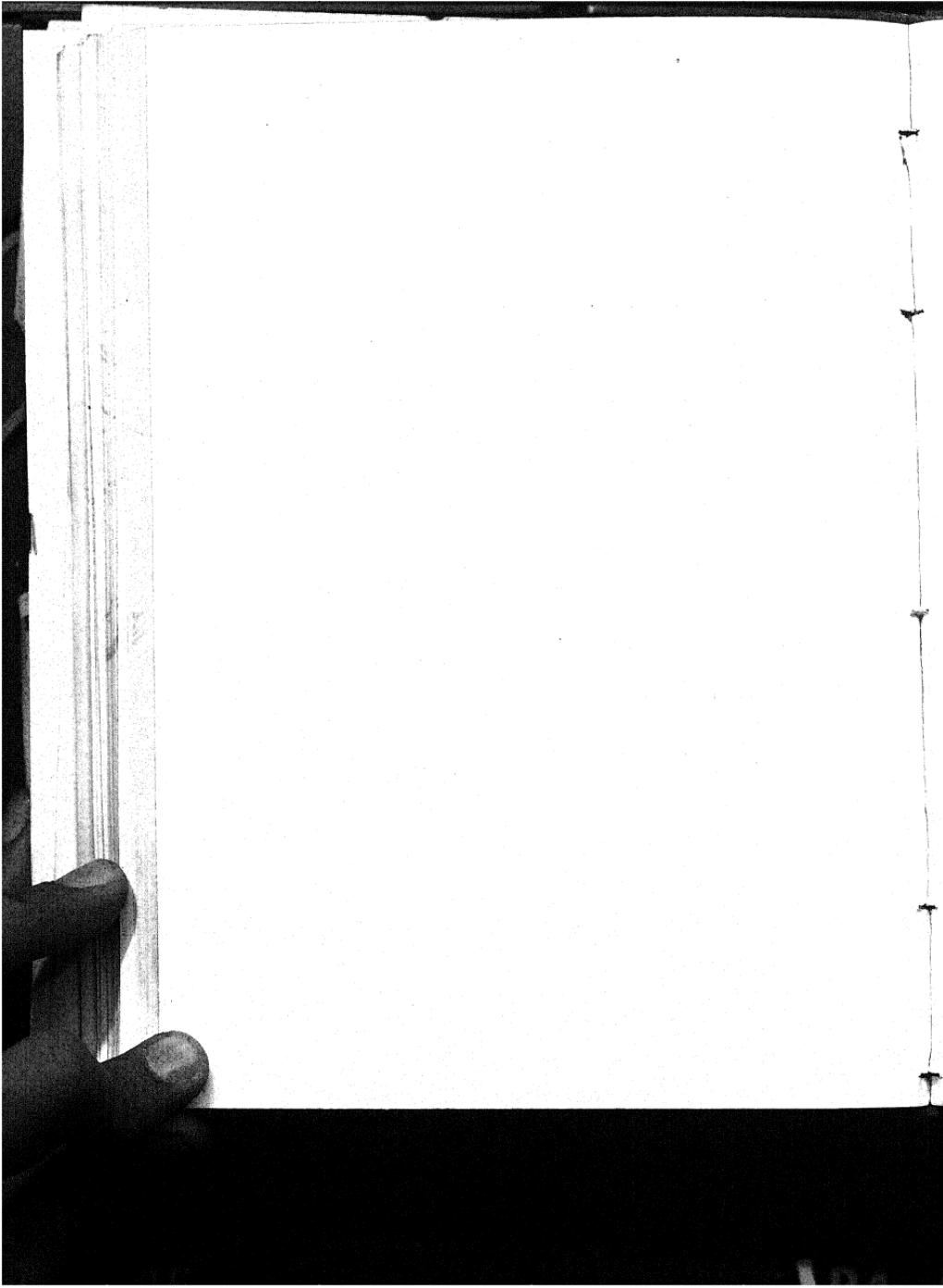
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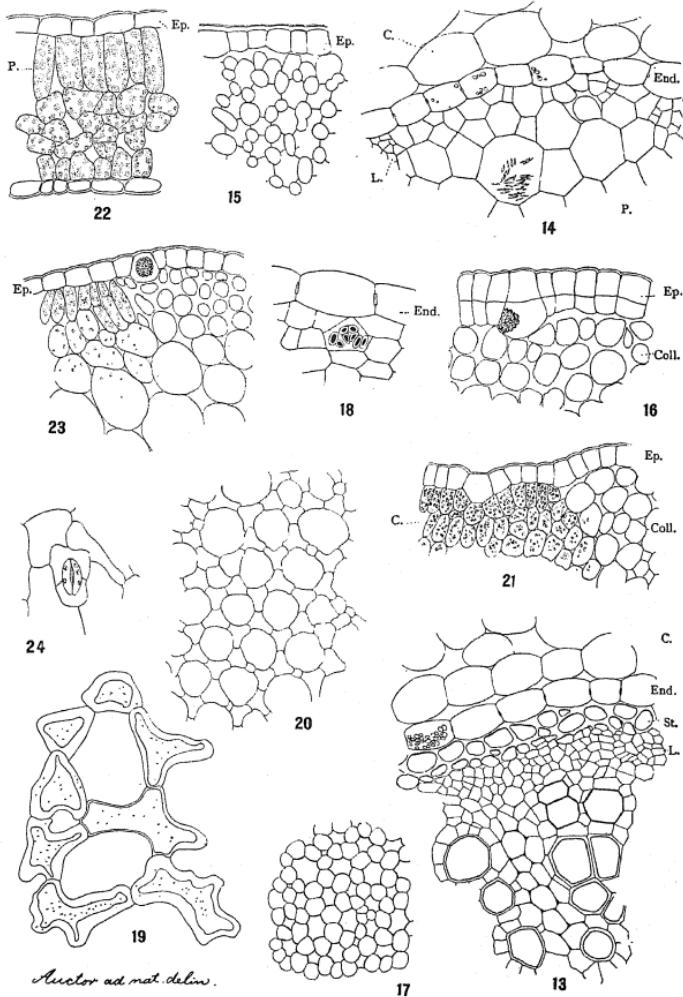
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HOLM on RUELLIA and DIANTHERA



seven steles, one in the center, six near the periphery; *s* and *s*, the four steles that fuse together in the node.

FIG. 10. Epidermis of upper face of leaf, showing a glandular hair and a stoma, viewed *en face*.  $\times 320$ .

FIG. 11. Epidermis and a glandular hair of leaf, transverse section.  $\times 320$ .

FIG. 12. Epidermis of lower face of leaf with a stoma.  $\times 320$ .

#### PLATE XII

FIG. 13. Transverse section of stem internode, showing part of peripheral stele: *c*, cortex; *end*, endodermis; *st*, stereome; *l*, leptome.  $\times 400$ .

FIG. 14. The innermost face of same stele (fig. 13), showing the continuous endodermis, some strands of leptome (*l*), and the large, thin-walled pith (*p*).  $\times 400$ .

FIG. 15. Transverse section of stem internode, showing the locally, very heavily thickened cell walls of the collenchyma inside epidermis (*ep*).  $\times 320$ .

FIG. 16. Transverse section of rhizome, showing beginning formation of cork in epidermis (*ep*).  $\times 320$ .

FIG. 17. Cortex of rhizome, transverse section.  $\times 75$ .

FIG. 18. Transverse section of rhizome, showing endodermis (*end*) and leptome with raphidines.  $\times 560$ .

FIG. 19. Transverse section of a node (submersed), showing the very thick-walled, branched cells of cortex.  $\times 240$ .

FIG. 20. Transverse section of an internode (submersed), showing the open cortex.  $\times 75$ .

FIG. 21. Transverse section of an aerial internode, showing collenchyma (*coll*) and palisades of cortex (*c*).  $\times 320$ .

FIG. 22. Transverse section of lateral portion of leaf, showing the ventral epidermis (*ep*) and the palisade tissue (*p*); below is the pneumatic tissue and dorsal epidermis.  $\times 240$ .

FIG. 23. Transverse section of leaf, showing palisades inside the dorsal epidermis (*ep*), and underneath the midrib.  $\times 240$ .

FIG. 24. Stoma of upper face of leaf.  $\times 320$ .

## THE IDENTITY OF MICROCYCAS CALOCOMA

OTIS W. CALDWELL AND C. F. BAKER

(WITH THREE FIGURES)

The important discoveries made in the Cycadales within the past decade led the first author to desire to secure material for a morphological study of *Microcycas*. During March and April of 1905 an abundance of staminate and carpellate cones as well as vegetative material of a small cycad said to be *Microcycas calocoma* were collected in Cuba. It was questioned immediately whether this material did not belong to *Zamia*. A careful comparison with herbarium specimens and an examination of literature soon led to the conclusion that it was *Zamia pumila*. Later investigation fully confirmed this conclusion and showed that it has been wrongly called *Microcycas calocoma*. Specimens of what is probably *Zamia pumila* have been issued several times from the New York Botanical Garden as *Microcycas calocoma*, the specimens coming from Madruga (Britton and Shafer 638 and 803) and from Matanzas.

In January and February 1907 a second attempt was made to locate and collect material of *Microcycas calocoma*, this excursion resulting more successfully than the first. In the higher regions of the Sierras of western Cuba a number of groups of the plant were found, also both staminate and carpellate cones. The confusion that exists in reference to the genus and the incompleteness of published accounts make desirable a somewhat full account of its bibliography and characters.

The earliest description discovered is that of MIQUEL in VAN HOUTTE'S *Fl. Serres et Jard.* (7:141. 1851-52), under the title "Sur une espèce nouvelle de *Zamia* des Indes occidentales, introduite dans l'establissement Van Houtte, à Gand." Although he gave this plant the name *Zamia calocoma*, in this same connection he suggested that there be made for it a new section of the genus, to be called *Microcycas*. In a note preliminary to the description MIQUEL says that this plant constitutes quite a distinct species, near *Z. tenuis*.

Willd., but differing from it in the form and number of the leaves. In this character MIQUEL says that *Z. calocoma* surpasses all other species of the genus, and "rappelle parfaitement le port d'un petit *Cycas revoluta*." MIQUEL's description is as follows:

*Zamia calocoma* Miq. Frondibus adultis glabris, pinnis densis numerosissimis, utrinque 65–70, coriaceis linearifalcatis subacutis integerrimis, marginibus leviter incurvis, utrinque tenere nervoso-striulatis, nitidis.—Sectio nova: *Microcycas*.

Adumbratio speciminis in Horto Amstelodamensi culti.

Truncus 10 poll. altus, basi 4½ crassus, subconicus, simplex vel bifidus, fere totus esquamatus, cortice hic illic lenticelloso-pustuloso, pallido, superne perulis coriaceis sensim rigescerentibus lanceolatis acuminatus ochraceo-griseo-tomentosis, glabrescentibus, circa frondium comam obvallatus. Frondes nunc undecim, erecto patentes. Stipes subtrigono-cylindraceus vel ferme cylindricus, antice non vel obsoletissime sulcatus, 3–4 poll. longus, calamus scriptorium crassus, juvenilis villo arachnideo tenui tectorum, adultus glaberrimus. Rhachis 1½–1¾ pedalis, rectiuscula vel flexuosa, subsemitereti-trigona, dorso valde convexa, antice interpinnas in angulum obtusum prominens, sulco utrinque pro inserendis pinnis haud profundo, apice in brevem mucronem incurvulum vulgo quidquam pilosum exserta. Foliola articulatione mox pallescente subtumidula inserta, senescentia seorsim decidua, media longiora quam superiora et inferiora (hinc tota frons lanceolata fere), omnia densa, pleraque alterna vel superiora subopposita aut opposita, horizontaliter patentia, saepe retrorsum arcuato-curvata, 3–11 in. distantia, infima paulo distantiora, linearia, rigidule coriacea, fere omnia falcata, supra saturate viridia, laevia, nitida, facie plana vel leviter canaliculata, subtus pallida, marginibus praesertim versus basim leviter recurva, utrinque tenere striata nervis parallelis in medio 12, versus, apicem foliolli saltē 5, saturate viridibus: omnia integerrima: media 4 poll. longa vel aliquid longiora, 1½–1¾ lin. lata. In plerisque frondibus utrinque 60–70 foliola numerantur.

In *Gartenflora* (6:16. pl. 186. fig. 30. 1857) REGEL mentions *Zamia calocoma*, but with no description. In 1857, however, he gives a brief description (Bull. Soc. Nat. Mosc. 1:191), based upon a specimen of *Zamia calocoma* which he says was brought from Cuba by CHAPPY. In addition to the characters given by MIQUEL, REGEL adds that the stem is one to two feet high and shows for a long time the markings left by the successive crowns. He also states that dormant buds appear on the stem. In closing he says that this plant varies so widely from any *Zamia* that it may constitute a new genus, and that he hoped his specimen would soon flower and thereby enable him to decide the question.

GRISEBACH (Cat. Pl. Cub. 217. 1866) refers to a further collection

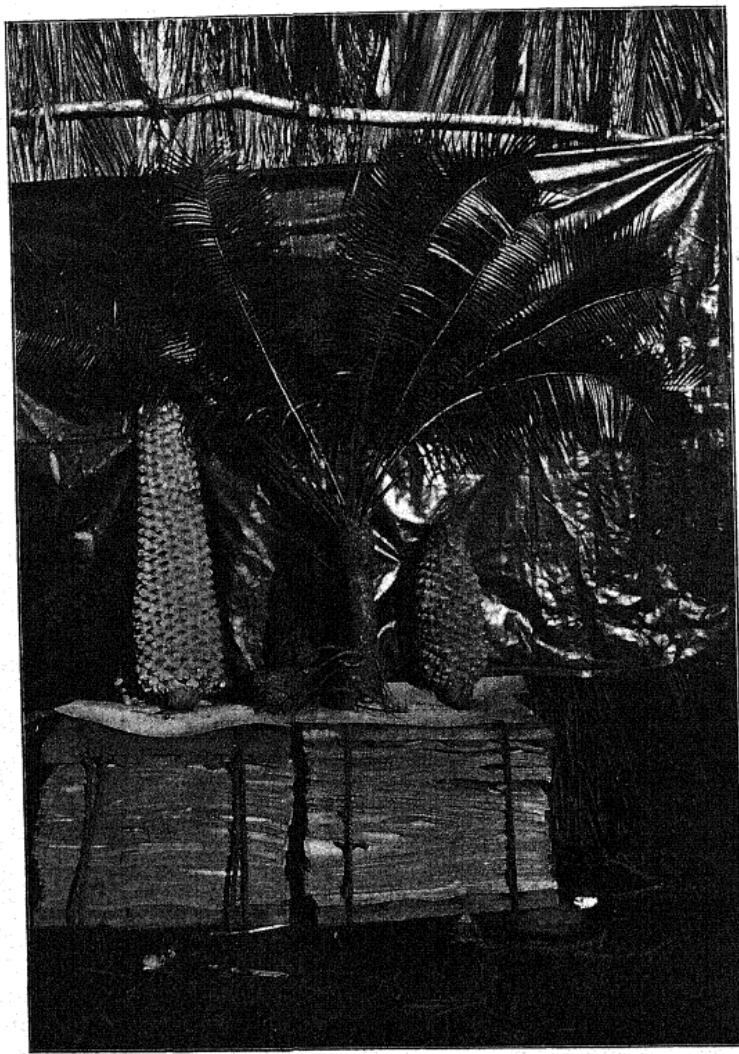


FIG. 1.—A young plant, two ovulate cones, one staminate cone (standing at right of left-hand cone), and terminal bud (at base of staminate cone); comparative sizes are shown by herbarium driers used as support for specimens.

by WRIGHT as follows: “*Zamia calocoma* Miq. (*Wr.* 3193: foliola pleraque 8" longa 4" lata; squamis ad *Z. Brongartii* Miq. accedit).”

In DE CANDOLLE’S *Prodromus* (16<sup>2</sup>:538. 1868) there appears the first full account of *Microcycas* as a distinct genus, the description being based upon previous descriptions and upon material collected by WRIGHT. From this we make the following excerpts:

Squamae strobili masculi planae, imbricatae? crassiusculae, a basi ad medium partem cuneatae et subtus crebre loculigerae, a medio ovatae, steriles. Squamae strobili feminei laxe juxtaposita, axi perpendicularares, superiore majore parte drupam irregulariter sulcatam simulante, nempe ellipsoideo-costatae et sulcatae, extremitate truncata vel obtusa, inaequaliter (in sicco?) bullata, basi in stipitem brevem angustata, stipite utrinque in apice semen ellipsoideum sessile gerente.—Arbuscula Antillana; trunco humili . . . nunc fructificatione cognita, genus inter Dioon et Zamiam appetet, a priore diversum foliolis basi segregatis et articulatis, squamis femineis pinguibus, apice contundatis et seminibus ut vulgo sessilibus; a Zamia, squamis masculis planis, non peltatis, femineis pelta multo longiore.

Under the description of the species (*M. calocoma* (Miq.) A.D.C.) the following statements appear:

Strobili masc. forma mihi ignota. Squamae (segregatae) 18–20 mill. longae, 9–10 mill. medio latae; pars fertili centim. longa, supra glabra, subtus undique loculis vestita; pars sterili cinereo-tomentosa, supra in nervum crassum medio inflata. Strobilus femin. 2 decim. longus, cylindricus? (in specimine sectus), pedunculo cinereo-tomentoso brevi? suffultus. Squamae 2 cent. longae, stipite 4–5 mill. longo, reliqua pars 15 mill. longa, 9–10 mill. lata, costis irregularibus latere sup. magis convexo, apice aut, planiusculo subquadragulo, aut contundato-sinuoso, pallide pilis stellatis tomentoso.

BENTHAM and HOOKER (*Gen. Plant.* 3:447. 1880) give an incomplete and imperfect characterization of the genus, and incorrectly cite the name of the species as *cubensis*.

From the above descriptions it is clear that our material is *Microcycas calocoma*; but it is also evident that the specimens used as a basis of these descriptions were far from being fully representative. There are two specimens of *Wright* 3193 in the Wright-Sauvalle herbarium, one of which possesses a young fruit. This is the number cited and described by DE CANDOLLE, and is identical with our own collections.

The following description is based upon our material:

Stem 25<sup>cm</sup> to 10<sup>m</sup> high, 1–6<sup>dm</sup> in diameter, often branched; bark

smooth or ridged, usually with conspicuous leaf scars; cross-section showing a thick cortex, a single vascular cylinder, and pinkish, brittle, starch-bearing pith: leaves 0.6 to 1<sup>m</sup> long, 6-40 in the crown; petioles

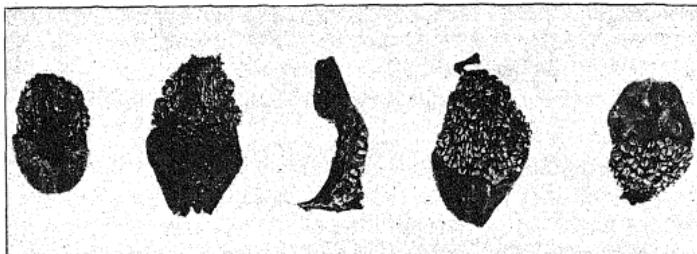


FIG. 2.—Staminate sporophylls; the two at the left show the adaxial surface, the two at the right the abaxial surface, the middle one a lateral view.

1<sup>dm</sup> long, terete, with shield-like base; leaflets 50-80 pairs, 8-12<sup>cm</sup> long, opposite or alternate, attached by an entire calloused base, finely villous when young, glabrous and glistening when mature, bright

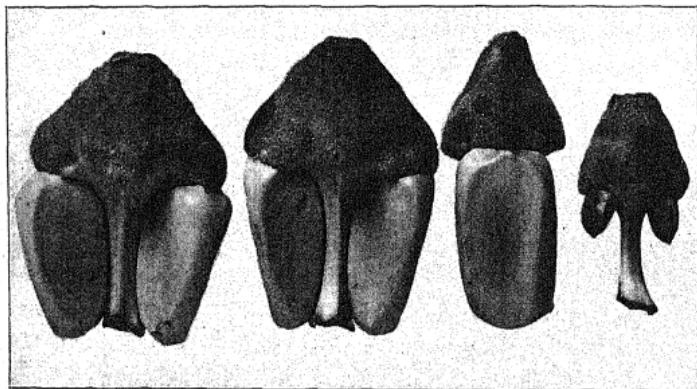


FIG. 3.—Ovulate sporophylls, showing (beginning at the left) abaxial view, adaxial view, lateral view, and abortive ovules.

green, with 15-20 dichotomously branching veins (reduced to 6-8 at the tip), bluntly pointed, tips turning inward slightly, margin slightly revolute: bud scales 4-6<sup>cm</sup> long, 1.5-2<sup>cm</sup> wide, often twisted at tip,

densely tomentose (except on lower half of inner face) with long gray hairs: staminate cone 25–30<sup>cm</sup> long, 5–8<sup>cm</sup> in diameter, cylindrical, on a short densely tomentose peduncle; sporophyll 2–2.5<sup>cm</sup> long, 1.5–2<sup>cm</sup> wide, the basal two-thirds of the abaxial surface densely covered by sporangia, the apical third gray-tomentose and prominently ridged along the median line, obtusely pointed: ovulate cone 50–70<sup>cm</sup> long, 13–16<sup>cm</sup> in diameter, cylindrical, slightly tapering from base to tip, obtusely pointed, on a short densely tomentose peduncle which together with the base of the cone is covered by densely tomentose scales 6–10<sup>cm</sup> long; sporophyll 4.5–5.5<sup>cm</sup> long, 3–4<sup>cm</sup> wide, 2–3<sup>cm</sup> thick, with stalk 3–3.5<sup>cm</sup> long and perpendicular to the axis of the cone, the outer portion quite convex, usually four-ridged, the apex blunt, sub-quadrangular, and sometimes furrowed, the entire outer portion of the sporophyll covered by a dense mass of closely appressed grayish hairs; ovules two, pink, 3.25–3.5<sup>cm</sup> long, 1.25–1.75<sup>cm</sup> thick (*figs. 1–3*).

STATE NORMAL SCHOOL, CHARLESTON, ILL.

ESTACIÓN AGRONÓMICA, SANTIAGO DE LAS VEGAS, CUBA

## BRIEFER ARTICLES

LINNAEUS<sup>1</sup>

MAY 23, 1707—JANUARY 10, 1778

(WITH TWO PORTRAITS)

The history of scientific progress has been marked by a few epochs introduced by new and revolutionary ideas. After a long period of almost no advancement, science has suddenly been carried forward in a most remarkable way. Such epochs have been few, and few are the men who have continued to be held in highest esteem with the steady progress of modern science. Among them was one who rose from humble descent to become the leading naturalist of the eighteenth century, and whose name is still among those most revered wherever natural science is taught or cultivated—LINNAEUS. Now that the day is at hand when his two-hundredth anniversary is to be observed throughout the scientific world, a brief sketch of his life and work is appropriate.

Between Lindshult and Jonsboda, in Sweden, there stood formerly an old and very large linden tree from which two brothers, the sons of a peasant, INGEMAR SVENSSON, adopted the surname TILIANDER; from this same tree their sister's son, NILS INGEMARSSON, took the name LINNAEUS. NILS INGEMARSSON was a minister, and his first living was in Raashult, the parish of Stenbrohult in Smaaland. He married CHRISTINA BRODERSONIA, and their first-born child was a boy who was christened CARL LINNAEUS. From his early childhood this boy showed an unmistakable love for flowers. His father was quite interested in botany and took pride in making the little garden of the parsonage one of the richest in flowers and rare shrubs. At the age of about four years CARL accompanied his father to a meeting; it was a fine summer day, and his father gathered several plants which he demonstrated before the guests, showing the peculiar roots of some orchids, *Succisa*, *Tomentilla*, and others. This little lecture in botany made such an impression upon the boy that he afterward never gave his father any peace, but repeatedly asked for more information and never tired of learning the names of all the plants he could collect. This incident was the first revelation of his genius;

<sup>1</sup> The 200th anniversary of the birth of LINNAEUS is to be celebrated in many places during May, and the BOTANICAL GAZETTE wishes to share in honoring the memory of so notable a botanist. At its request this sketch has been prepared by Mr. THEO. HOLM, whose selection for the task is justified by his high esteem of the man and intimate knowledge of his life and work.—EDS.

and although he was "doomed to become a minister" while still in the cradle, he always remained faithful to his first impulse to become a student of nature.

As he grew older, this first impulse drove him farther and farther; he commenced to collect plants and insects, and in school he was soon dubbed "the little botanist." Some of his teachers, LANNERUS and ROTHMAN, took a special interest in him, and it was at the urgent request of ROTHMAN that his father abandoned the idea of having his son educated for a minister. LINNAEUS was then sent to Lund to study medicine, and there he had the good fortune to meet the famous STOBAEUS, who soon discovered his talent and aided him in every way to prosecute his studies. He showed him the large collections in the museum, which at that time was quite rich in minerals, shells, birds, and dried plants. Young LINNAEUS took a great interest in these collections and especially in the herbarium, and he very soon began to study the local flora, collecting and drying plants and mounting them.

From Lund he went to Upsala, where he knew he would find a larger library, a botanical garden, and more than all he would have an opportunity to attend the lectures of ROBERG and RUDBECK. On his arrival in Upsala (1728), he met CELSIUS, who treated him with the same fatherly benevolence as STOBAEUS. CELSIUS introduced LINNAEUS to RUDBECK, jr., an acquaintance who became of great importance to him. RUDBECK had himself visited Torneaa, Lapmark, and brought home extensive collections of plants, all of which were lost, however, by the great fire of Upsala (1707). While staying in the house of RUDBECK, LINNAEUS often listened to his narratives about this journey, and he soon felt a vivid desire to see that country and study its fauna and flora. Lapmark was at that time a *terra incognita*, and it was not so strange that the Royal Swedish Society accepted the proposition of CELSIUS and RUDBECK to furnish LINNAEUS with the necessary funds for making a journey to that country to make new collections.

LINNAEUS left for Lapmark in the month of May 1732, and, having made numerous excursions throughout the country, he returned to Upsala in the late fall of the same year. To LINNAEUS the journey became of the greatest importance. The arctic-alpine vegetation that prevails there left a deep impression upon him; his views became broadened; and he acquired great experience in field observation, not only in distinguishing species, but also in the geographic distribution of plants and animals. The results of this journey as they appeared in his *Flora Lapponica* laid the foundation of his fame. In the following year LINNAEUS visited the Swedish province

Dalarne, at the request of Baron REUTERHOLM, who wished him to explore that country as he had explored Lapmark.

While thus LINNAEUS had devoted much of his time to the prosecution of his studies in natural history—botany, zoology, and mineralogy—he managed, nevertheless, to finish his course in medicine. It became clear to him, however, that his prospects as a physician in Sweden were not very promising unless he took the degree of Doctor medicinae. It was then

that his friend JOHAN BROWALLIUS persuaded him to take this step, while his father-in-law, Dr. JOHAN MORAEUS, offered to pay his expenses. He selected the University of Harderwijk, a small town in Gelderland, Holland, which at that time was frequently visited by Swedish physicians. In the early spring of 1735 LINNAEUS left Sweden and arrived in Amsterdam June 2; thence he proceeded to Harderwijk, and having passed the preliminary examination he presented as his dissertation "Hypothesis nova de febrium intermittentium causa." Having proved his knowledge of medicine, LINNAEUS received the "diploma, golden ring, and silk-hat" June 23, 1735.

From Harderwijk LINNAEUS went to Amsterdam, where he visited



LINNAEUS in Lapland costume; from an oil painting by Hoffman in Amsterdam, 1737.

BURMAN, and thence he went to Leyden. His meeting with GRONOVRIUS, BOERHAVE, and CLIFFORD of Hartecamp secured him the unusual opportunity of obtaining an excellent position as naturalist, and also the opportunity of seeing the most famous museums and botanical gardens in Europe. It was at the suggestion and expense of CLIFFORD that LINNAEUS visited England and France, and so great was his admiration for LINNAEUS that he tried to persuade him to make Holland his home, but he returned to Sweden in May 1738.

It is really remarkable to note the good fortune that followed LINNAEUS wherever he went, in meeting congenial friends and in gaining their confidence and admiration. His strong personality and his enthusiasm for

natural science impressed everyone with whom he came in contact. His work in Holland resulted not only in the development of the large collections at Hartecamp, but also in the publication of some of his greatest works. Twelve important works were published during the two and a half years spent in Holland among which were the following: *Systema naturae*, *Musa Cliffortiana*, *Bibliotheca botanica*, *Fundamenta botanica*, *Flora Lapponica*, *Critica botanica*, *Hortus Cliffortianus*, *Genera plantarum*, and *Classes plantarum*.

On his return to Sweden, LINNAEUS was appointed surgeon of the admiralty and *Regius Botanicus*, and a few years afterward he was made Professor ordinarius at the University of Upsala (1741), and became the founder of the Museum Academicum. As a teacher, LINNAEUS showed an indefatigable interest in his pupils; by lectures and excursions in the field he taught them how to study nature, and it was under his patronage that several young men were sent to foreign countries to make collections. In this way, and also through his numerous friends among foreign botanists, LINNAEUS succeeded in accumulating an enormous mass of material for the completion of his work.

His aim to make natural science popular was crowned with success. It is difficult to find a whole series of publications by one man, in which the materials of natural science are brought together and presented so intelligibly as in the works of LINNAEUS. The simplicity that characterized his life was also expressed in his scientific methods. His ability to describe species of animals and plants in a very few words was unexcelled. He was the first to establish a system by which all plants might be readily classified, and although purely artificial this system has proved to be very useful, and is to some extent really a part of what we call now the natural system; and the terminology which he proposed is the one that is followed today.

The credit for having introduced binomial nomenclature as a "general principle" also belongs to LINNAEUS. He was actually the founder of plant morphology and plant geography, which may be seen from the following dissertations: *Prolepsis plantarum*, *Sponsalia plantarum*, *Gemmae arborum*, and from the introductions to his *Flora Lapponica*, *Hortus Upsaliensis*, etc. He studied animals and plants as living organisms, as members of a great living world, and he outlined in a remarkably simple way the distinction that may be observed between species and varieties. His greatest talent consisted in his unique power to grasp nature as a whole and to classify; but to classify in the spirit of LINNAEUS is something more than is understood by classification in modern times. He was com-

peled to invent a terminology, and to establish rules for describing and naming species as well as genera; and even if the system which he established was purely artificial, he indicated by his "Ordines naturales" that a natural system is the supreme end of systematic work.



LINNAEUS at the age of 67; from a painting by Per Krafft.

Upsala. A beautiful monument was erected on his grave, and the brief inscription shows how much he was appreciated by his friends and pupils. It is as follows:

Carolo a Linné  
botanicorum principi.  
Amici et discipuli.  
MDCCXCVIII.

To his last days he preserved the friendly and simple nature that had marked him from his youth. Very characteristic of him was his susceptibility to impressions and his indomitable energy. During the long period of active life he always declined to enter into discussion with those who opposed him; and he was accustomed to say: "If I have erred, nothing will be gained; but if I am right, I shall be so as long as nature exists." He was a very religious man and a firm believer in the inevitable Nemesis.

He died January 10, 1778, and was buried in the old cathedral of

### THE ORGANIZATION OF THE ECOLOGICAL INVESTIGATION OF THE PHYSIOLOGICAL LIFE-HISTORIES OF PLANTS<sup>1</sup>

Upon several occasions I have expressed the belief that the first need of plant ecology is the determination of the physiological life-histories of the forms concerned. I propose now to explain more fully what I mean.

First, it is essential to note our changing conception of ecology. It is not long since we sought a use for every detail of plant structure and habit, and, correlatively, assumed that these had been developed in some kind of causal touch with the environmental conditions they were supposed exactly to fit. This was causal, or historical, adaptation, and it was the logical product of the Darwinian assumption of a struggle for existence so keen as to make even minutiae determinative of the survival or extinction of whole organisms, and through them of entire species. Now, partly because of the work of DE VRIES, but partly, I venture to assert, because of the studies of ecologists themselves, we are ceasing to require utility in details of structure and habit, and are coming to believe that such details may be determined by factors wholly independent of the immediate environment. This newer view does not exclude the possibility of causal or historical adaptation in details, but it accepts the probability of a very different origin and meaning of those details. And this different non-causal origin and meaning of the relation between the details of structure-habit and environment may be either of these two. First, the adaptation may be real but a matter of accident or coincidence, the details arising from internal or other irrelative causes and finding their appropriate environment by a process of sifting. Second, the details, however arisen, may have no positive relationships whatever with the environment, but, not happening to run counter to any very potent feature of that environment, they exist by toleration. In brief, while adaptation as a broad, a general, a generic matter is a reality and rests upon a certain historical or causative basis, adaptation as an exact, a minute, a specific matter is quite different; it may be causative, but it is more often coincidental or tolerative. The ecology of the future will be, primarily, not a search for utilities, but an analysis of meanings.

I emphasize this matter thus fully because I believe that in the phase of ecology of greatest present-day interest, viz., physiognomic ecology, the study of the factors determining the features of vegetation, it will prove most illuminating. A great deal of our supposed adaptation of vegetation

<sup>1</sup> Read before the Botanical Society of America at the New York meeting, December 29, 1906.

to habitat is, I have no doubt, not adaptation in the old sense at all, but is a sifting-in of forms whose characters have been elsewhere and otherwise determined, but which happen to match, either by coincidence or by toleration, with the physical conditions prevailing in that habitat. So far do I consider this true that I think our best working hypothesis in this field is this: any plant stands where it does for the reason that the physical demands made by the structure and habit it happens to possess overlap in some degree the physical conditions prevailing in that place, and the better they match the more nearly does the plant find its optimum, and the worse they match the more slender is the hold of the plant upon that place. Now the study and measurement of the physical factors of environments is steadily progressing; it behooves us to study and measure the physical demands of the plants in order that we may be able to compare the one with the other. This is the crux of what I mean by the need for physiological life-histories of plants.

I come now to the practical part of the subject. I assume that the plant is a bundle of physical needs, and that our aim is to determine what and how great these are. Then these things are needful:

1. We must devise methods and instruments, for the most part autographic, for measuring and recording each physiological process. This, indeed, we are doing, but must do more and better.

2. We must develop a system of standard units for each physiological process, such as will permit their exact expression and their comparison. We already measure photosynthesis in grams per square meter of surface per hour, and are approximating to similar definite units for respiration and transpiration. We must develop similar systems for the other processes. Further, we must develop methods of graphic expression of such data in order that they may be readily matched against the graphic results supplied by study of the environments. In most cases these will doubtless work themselves out in the form of minimum-optimum-maximum curves, but it will not be enough to make these curves represent means or averages; they must express the range of frequency from the mean, which can be done by means of shaded or penumbrate graphs, in which the mean forms the dark center and the shading merges off thence to the extremes.

3. We must not confine our measurements to those invisible processes in the protoplasm which we commonly associate with physiology, but must include structure which is simply an external and visible manifestation of physiological operations, a tool which physiology forges as an aid in its processes. Does a plant produce a long tap-root? That is because, for reason sufficient unto itself even though unknown to us, the protoplasm

of that plant extends itself deep into the ground; the structure is a visible physiological result, and is to be measured and expressed like the other physiological processes.

4. We must especially learn to distinguish and to express to what degree the various physiological processes are plastic to the action of stimuli, and to what extent are limited by heredity.

5. We must classify the physiological processes for study. Ecologically speaking there are four critical periods in the life-cycle of a plant. These are (*a*) the germination of seed or spore, (*b*) the orientation of the seedling whereby the plant gains a grip upon its immediate surroundings, (*c*) the expansion of the adult, and (*d*) flowering and fruiting, or sporification. It is not enough that the plant can match an environment in three of these; it must match in all four. I have no doubt dissemination brings regularly into a certain habitat some plants which can germinate their seeds there, flourish as adults, flower and fruit, but in which the seedling stem and leaves cannot stand the light and heat to which they are exposed, or in which the seedling root cannot reach the permanent water supply, and accordingly that kind of plant cannot exist in that habitat. I think it is some such determinant which brings it to pass that, of all the trees whose seeds are brought upon the Miscou sandplains from the neighboring upland, not one can exist there except the white spruce. I think many peculiarities of ecological distribution are determined by just such single seemingly insignificant causes. But to explain such cases we must know the plants, and must know them throughout their life-cycles; and this is the essence of a knowledge of physiological life-histories.

The particulars of a classification of physiological-ecological processes can be worked out only by experience; but following are some of the more important topics:

A. Germination of the seed:

Amount of water requisite, least and best, and duration thereof.

Temperature requisite, minimum, optimum, and maximum, and rate under each.

Light, whether necessary, injurious, or indifferent.

Quality of water requisite; how much of salts it may contain.

Free oxygen requisite, or can it germinate under water?

Germinates on the surface or needs planting, and why?

B. The seedling life:

Root system; ordinary form and size and limits of irritable adjustment.

Stem and leaf system, ordinary form and size, and limits of irritable adjustment.

Temperature conditions.

- Water, amount and quality requisite.
- Light, amount requisite for photosynthetic work, least, best, and most.
- How far carried by stored food, and when its own nutrition begins.
- C. Adult life:
  - Demand for water, and interadjustment of absorption, transfer, and transpiration.
  - Demand for the important minerals.
  - Dependence upon mycorrhiza or other organic aids.
  - Form and size of vegetative parts, and plasticity thereof.
  - Temperature demands.
  - Light demands.
- D. Flowering and fruiting (sporification):
  - Conditions determining beginning of reproduction.
  - Vegetative powers of reproduction.
  - Arrangements for uniting the sexes, and extent of dependence thereon.
  - Duration of vitality, and the resting period of the seed.
  - Dissemination conditions.

—W. F. GANONG, *Smith College, Northampton, Mass.*

## CURRENT LITERATURE

### MINOR NOTICES

Javanese woods.—Under the direction of Dr. J. W. MOLL, of Groningen, H. H. JANSSONIUS has undertaken to describe in detail the structure of a large collection of the woods of Java,<sup>1</sup> which was made by KOORDERS in connection with his undertaking to prepare for the Buitenzorg Garden a forest flora. KOORDERS marked 4000 individual trees, many of them in the primitive forest, and from these and others collected 15,000 specimens, which form the basis of the work, *Flora arborea javanica*, by KOORDERS and VALETON. The collection of woods comprises 2400 numbers, representing 1070 species, 380 genera, and 80 families. Provision is made for the complete identification of this material with corresponding herbarium material which has been or will be distributed. The descriptive part of this work comprises, under each species, references to the literature (an enormously repetitious method), an account or list of the material, names of the preparations (whether cross, radial, or tangential sections, or macerations), a list of the reagents employed (why not also one of the tools used?), and finally the micrography, including the topography (which is usually illustrated) and a detailed description of the elements. The utmost pains is evident in the attention to every detail of organization and of execution; and if such an enormous accumulation of minutiae is of value to anybody he will certainly find this work a mine of them. It is being issued with the support of the Colonial Office. Professor MOLL desires authors whose work has dealt with the anatomy of woods to send him separates.—C. R. B.

*Flora of Columbia, Missouri*.—The second number of the science series of "The University of Missouri Studies" is a handsome and bulky volume on the flora of Columbia and vicinity, by F. P. DANIELS.<sup>2</sup> It represents the results of the intensive study of a restricted flora for five or six years, and consists of two parts bearing the titles "Ecology" and "Flora."

Under "Ecology" (pp. 75) "the attempt has been made to indicate the social relationships of the flora; to determine the external factors of these associations; and to ascertain the physiological adaptations of the plants themselves to their environment." The account of plant associations and their distribution, and of the composition of the flora, is to be regarded as a finished one; but the experi-

<sup>1</sup> MOLL, J. W., and H. H. JANSSONIUS, Mikrographie des Holzes der auf Java vorkommenden Baumarten. Erste Lieferung. 8vo. pp. 368. figs. 44. Leiden: E. J. Brill. 1906. M6.

<sup>2</sup> DANIELS, F. P., The flora of Columbia, Missouri, and vicinity; an ecological and systematic study of a local flora. pp. vi+319. 1907.

mental work necessary to establish the conclusions in reference to factors and adaptations remains to be done.

Under "Flora" (pp. 200), 1058 native and introduced species are enumerated, representing 435 genera and 101 families. Attention is called to the fact that the most striking feature of the flora is the large number of Gramineae, Leguminosae, and Compositae, and the almost complete absence of Coniferae, Orchidaceae, Ericaceae, and Gentianaceae. New species are described under Physocarpus (2) and Vernonia (9), and a synopsis of the latter genus appears in an appendix. Several appendices, an elaborate summary, and a very full index show a real consideration for the convenience of the reader.—J. M. C.

**Irritability.**—FITTING prepared, at the request of the editor of *Ergebnisse der Physiologie* in 1905 an essay entitled *Das Vorkommen von Reizleitungsvorgängen bei den Pflanzen und die Methoden zu ihrem Nachweise*, and in 1906 for the same work a second, *Der Ablauf der Reizleitungsvorgänge bei den Pflanzen*. These essays are synoptical in their character and include extensive references to the literature of the subject, with the intention that these summaries might be useful to other physiologists as a source of information and to plant physiologists might serve as a starting-point for further investigations. Inasmuch as the work in which they appeared has a rather limited circulation, especially among botanists, it seemed wise to the author to publish the two papers also as an independent physiological monograph; and in this form they do indeed make a small volume,<sup>3</sup> which can be had at a low price and thus becomes readily available. This work puts the facts of the matter succinctly, discusses the theories of conduction critically though briefly, and therefore is likely to serve the very purpose which the author had in mind. The original separate publication has involved the retention of the bibliography in two lists, largely duplicates; but this is a small inconvenience in view of the larger service.—C. R. B.

**German south-polar expedition.**—Of the sumptuous quarto volumes reporting the results of this expedition in 1901–3, the seventh is devoted to botany. In the first part, which has just been published,<sup>4</sup> HENNINGS reports upon the Fungi, describing over 30 new species (2 plates); ZAHLBRUCKNER treats the lichens, among which he finds 17 new species and a new genus, *Steinera* (Lichenaceae), and takes occasion to describe more fully a number of little-known species (3 plates); SCHIFFNER lists the liverworts (of which about 40 were previously known), describing 3 new species and a new form out of the 14 collected (1 plate); BROTH-ERUS found in the collections 61 mosses, of which 14 were new (3 plates); SCHENCK reports upon the vascular plants of each island, but finds no new species.

<sup>3</sup> FITTING, HANS, Die Reizleitungsvorgänge bei den Pflanzen. Imp. 8vo. pp. xvi + 157. figs. 15. Wiesbaden: J. F. Bergmann. 1907. M. 3.60.

<sup>4</sup> Deutsche Südpolar-Expedition 1901–1903, im Auftrage des Reichsamtes des Innern, herausgegeben von ERICH VON DRYGALSKI, Leiter der Expedition. VIII Band, Botanik, Heft 1. 4to. pp. 176. pls. 10. figs. 25. Berlin: Georg Reimer. 1906. M. 40.

About one-third of the volume is devoted to a discussion by Dr. E. WIRTH of the ecological aspects of the vegetation, and this is illustrated by unusually fine figures and 10 plates (half-tones and photogravures), showing the formations and some individual plants. The annual course of vegetation on Kerguelen is also treated at some length.—C. R. B.

**Errera Botanical Institute.**—There has come to hand the second volume of papers from this institute, two volumes of which, published earlier, were noticed in this journal recently.<sup>5</sup> This volume<sup>6</sup> contains several papers on nitrates and ammoniacal salts, especially in relation to yeasts and other fungi and to root-tuberles, by LAURENT and by MARCHAL; some brief notes on microscopical and bacteriological technique by ERRERA and DE WEVRE; a considerable number of papers on the occurrence, localization, and significance of alkaloids in plants by ERRERA, CLAUTRIAUX, MAISTRIAUX, MOLLE, DE WEVRE, DE WILDEMAN and DE DROOG; several papers on proteids by ERRERA, MARCHAL, CLAUTRIAUX, and DE WEVRE. All these papers have been published before, dating from 1881 to 1904, but mostly between 1888 and 1895. The volume closes with an annotated bibliography (41 pages) on alkaloids, glucosides, and tannins, etc., brought together by ERRERA and not previously published.—C. R. B.

**British vascular plants.**—MESSRS. JAMES BRITTEN and A. B. RENDLE have compiled a list of British seed-plants and ferns<sup>7</sup> in accordance with the international rules of nomenclature adopted by the Vienna Congress. The list is most timely and will prove a great convenience to students of the British flora. The British systematists are now in a condition of most enviable uniformity as to nomenclature, evidently having taken part in the Vienna Congress in the expectation of abiding by its decisions, and giving up, for the sake of harmony, even so cherished a custom as using the specific name first employed in the genus accepted. If our own flora were not so extensive, and if our own systematists were content to waive individual opinions that did not happen to be accepted by the Congress, such a working-list might be prepared for this country.—J. M. C.

#### NOTES FOR STUDENTS

**Items of taxonomic interest.**—L. M. UNDERWOOD (*Bull. Torr. Bot. Club* 33: 591-605. 1906) has revised the American species of *Stenochlaena*, 12 being recognized and 3 new ones described.—W. H. BLANCHARD (*Torreya* 7: 7, 8. 1907) has described a new *Rubus* (blackberry) from Mass. and R. I.—A. W. EVANS (*Bryologist* 10: 24-30. 1907) discusses and approves the reasons assigned by

<sup>5</sup> *BOT. GAZETTE* 43: 215. 1907.

<sup>6</sup> Recueil de l'Institut Botanique (Université de Bruxelles) publié par L. ERRERA. Tome II. Imp. 8vo. pp. xii+415. pls. 4 figs. 3. Bruxelles: Henri Lamertin. 1906. fr. 20.

<sup>7</sup> List of British seed-plants and ferns. Department of Botany, British Museum (Natural History). pp. 40. Published by the Trustees of the British Museum. 4d.

LEVIER for retaining the genera *Calypogeia* and *Gongylanthus* as against *Kantia* and *Cincinnulus*, gives the synonymy of *C. fissa*, mentions the other N. Am. species, and proposes a new combination (*C. portoricensis*) for a West Indian species.—F. LAMSON-SCRIBNER (*Rhodora* 9:17-23. 1907) has presented the eastern species of *Muhlenbergia*, suggesting a somewhat new classification and segregating several sub-species and one new species.—M. L. FERNALD (*idem* 23-25) has described the alpine *Rhinanthus* of Quebec and New Hampshire as a new species (*R. oblongifolius*).—W. W. EGGLESTON (*Torreya* 7:35, 36. 1907) has described 2 new species of *Crataegus*, one from Vermont, the other Mexican.—P. A. RYDBERG (*Bull. Torr. Bot. Club* 34:35-50. 1907), in his 17th "Studies of the Rocky Mountain flora," has described new species in *Pedicularis*, *Adenostegia*, *Castilleja* (13), *Lupinus* (10), *Trifolium* (4), *Tium*, *Hamosa*, *Xylophacos*, and *Homalobus* (2).—L. DIELS (*Bot. Jahrb.* 39:469-486. 1907), in a synopsis of African *Anonaceae*, describes *Tetraستemma* as a new genus.—A. ENGLER (*idem* 573-580) has published *Sloetiopsis* (*Moraceae*) and *Pierreodendron* (*Simarubaceae*) as new African genera.—A. BRAND (*Pflanzenreich*, part 27. 1907), in a monograph of *Polemoniaceae*, recognizes 12 genera and 277 species, and describes new species in *Cantua* (2), *Polemonium* (3), *Phlox* (9), *Gilia* (12), *Navarretia* (7), and *Langloisia*.—W. BOTTING HEMSLEY (*Annals of Botany* 21:71-77. pls. 9-10. 1907) has described a new genus (*Seychellaria*) of *Triuridaceae* from the Seychelles.—CARDOT and THERIOT (*Univ. of California Publications, Bot.* 2:297-308. 1906), in a list of Alaskan mosses collected by W. A. SETCHELL and others in 1889, describe and illustrate new species in *Orthotrichum*, *Bryum* (2), and *Hypnum*.—H. CHRIST (*Bull. Herb. Boiss. II*. 7:257-274. 1907), in presenting the ferns of his *Costa Rican Flora*, describes 17 new species; and also (*idem* 275, 276) describes a new fern genus (*Lathyropteris*) from Madagascar, with characters intermediate between those of *Pellaea* and *Pteris*.—E. LEMMERMAN (*Bot. Jahrb.* 38:343-382. 1907), in an account of the algal flora of the Chatham Islands, describes a new genus (*Dermocarpella*) of *Chamaesiphoniaceae*.—G. W. WILSON (*Bull. Torr. Bot. Club* 34:61-84. figs. 10. 1907), in the first paper of a series entitled "Studies in N. Am. Peronosporales," presents the genus *Albugo*, gives lists of hosts, recognizes 13 species, and describes one as new.—Miss G. S. BURLINGHAM (*idem* 85-95) has described 6 new species of *Lactarius* from Vermont.—C. H. PECK (*idem* 97-104) has described 20 new species of fungi, in 18 genera.—J. M. C.

**Biometric studies.**—FAWCETT<sup>8</sup> has studied the variation in number of ray-flowers in the following Compositae: *Anthemis Cotula* from seven localities, *Achillea Millefolium* from four localities, and *Senecio triangularis*, *Aster adscendens*, and *Erigeron salsuginosus*, each from one locality. The results are not well discussed, but the data are given in tables and also represented by curves which show the following facts of interest: (a) Only in the case of *Anthemis* and *Achillea* were the numbers sufficient to give dependable results. (b) Both of these give

<sup>8</sup> FAWCETT, H. S., Variation in ray flowers of *Anthemis Cotula* and other Composites. *Proc. Iowa Acad. Sci.* 1905:55-58. pls. 12-20. 1905.

strongly monomodal curves, the former with the mode at 13, the latter with the mode at 5, thus supporting LUDWIG's view as to the prevalence of the terms of the Fibonacci series as modal numbers in the variation curves for ray-flowers. (c) Individual variation is shown in several tables, including about 70 individuals of *Anthemis* and 40 individuals of *Achillea*. These show that considerable differences occur, but so far as can be seen each habitat studied shows about the same series of individual variabilities. (d) Collections of the same species from different localities show little place-variation except in one instance. The number of rays in the heads of *Achillea* are only four in a much larger percentage of the heads at Ames, Iowa, than at three other localities from which material was secured. (e) Collections of *Anthemis* in a single habitat near Ames, in August and again in September, show a somewhat higher average number of rays at the time of the earlier collection, in this agreeing with results reached by the reviewer in the genus *Aster*. The modal number was 13 in both collections. It is stated that greater variability was observed in the ray-flowers of *Anthemis* growing in rich soil than elsewhere, but the data upon which this statement rests do not appear in the tables.

The number of seeds of the lotus, *Nelumbium luteum*, as it occurred in the western part of Lake Erie in 1902, has been studied statistically by PEARL,<sup>9</sup> who finds that the distribution of the variates gives a good example of the normal curve with the following constants: average  $24.874 \pm 0.078$ , standard deviation  $4.339 \pm 0.055$ , coefficient of variation  $17.445 \pm 0.162$ . A complete analysis of the curve is made and its departures from the theoretical are accounted for as due to random sampling. An interesting numerical relation bearing upon the distribution of fecundity is pointed out. It appears that capsules bearing fewer than the median number of seeds produce only 42.96 per cent. of the total number of seeds, while the remaining 57.04 per cent. are borne by plants which severally produce more than the median number. This fact is conditioned by the normal form of the curve. The maintaining of the normal curve would seem to require either that the inheritance of seed-number is inappreciable or that there is unequal elimination of the seeds coming from large and from small heads.—G. H. SHULL.

**Development of the seed of Alsinoideae.**—Miss GIBBS,<sup>10</sup> in working over the development and germination of the seed in the subfamily Alsinoideae, of the Caryophyllaceae, has shown that in these features there is a marked uniformity in this group, with evidences of a progressive specialization showing a trend from the Sperguleae to the Alsineae. The latter is indicated especially by an increase in size, and presumably in efficiency as an absorbing organ, of the suspensor; and also by the increased specialization of the inner layer of the outer integument, which is believed to serve as a water jacket in the developing seed. The structure

<sup>9</sup> PEARL, R., Variation in the number of the seeds of lotus. Amer. Nat. 40:757-768. figs. 4. 1906.

<sup>10</sup> GIBBS, L. S., Notes on the development and structure of the seed in the Alsinoideae. Annals of Botany 21:25-55. pls. 5-6. figs. 4. 1907.

of the chalazal region of the seed was studied in some detail, and the attempt is made to correlate the structure with the functions of this region in conveying food to the young seed after the cutinization of its coats, and in providing for the necessary gaseous interchange with the seed.

The embryo-sac is developed, without division to tapetum or megasporae, from a hypodermal cell of the ovule. The antipodal cells are evanescent, and the multinucleate endosperm-sac grows to occupy the whole peripheral region of the campylotropous ovule, and is finally filled by the strongly curved embryo. The endosperm is a single layer of cells throughout, though much thicker in the micropylar cap of endosperm which covers the radicle in the mature seed. This part of the endosperm seems the more important, and evidently serves the part of a digesting and absorbing structure in transferring food material from the nucellus to the embryo during its later intraseminal development and the early stages of germination. This function is indicated by the position of the endosperm, by the nature of its cell contents, and by the fact that seeds from which the endosperm has been removed do not germinate.

This work of Miss GIBBS adds another series of perisperm-containing seeds to those in which JOHNSON<sup>12</sup> and LEWIS<sup>13</sup> have shown that the endosperm is not for the storage of food for the embryo, but serves to digest and pass on to the embryo, before and during germination, the starch stored in nucellar tissue.—DUNCAN S. JOHNSON.

The structure of *Trigonocarpus*.—SCOTT and MASLEN<sup>13</sup> have been investigating the structure of certain species of *Trigonocarpus*, a common seed-genus of the British Coal-measures. The species discussed are *T. Parkinsoni*, which name replaces the more familiar *T. olivaeforme*, and *T. Oliveri*, a new species. The general oval form of the winged body of the seed and its very prominent micropylar beak are the familiar features. The testa is distinctly cycadean in character, the outer fleshy and the stony layers being very evident, while there are traces of the inner fleshy layer. The micropylar beak is often as long as the body of the seed, and in some specimens twice as long, and is peculiar in bearing two broad wings, whose real nature is problematical. The nucellus and testa are free throughout, a feature, presumably an ancient one, belonging to certain paleozoic seeds. The vascular system of the seed is in general that described by OLIVER for *Stephanospermum*, namely, an outer system running in the outer fleshy layer, and an inner system traversing the nucellus. This nucellar system in *Trigonocarpus* is quite conspicuous, forming a "practically continuous sheath" in the lower part of the nucellus, and traceable through the whole length of the nucellus, almost to the base of the pollen chamber. The pollen chamber is rela-

<sup>12</sup> BOT. GAZETTE 34:321-340. pls. 9, 10. 1902.

<sup>13</sup> BOT. GAZETTE 40:79. 1905.

<sup>13</sup> SCOTT, D. H., and MASLEN, ARTHUR J., The structure of the paleozoic seeds *Trigonocarpus Parkinsoni* Brongniart and *Trigonocarpus Oliveri*, sp. nov. Part I. Annals of Botany 21:89-134. pls. II-14. 1907.

tively small and has not been found to contain pollen grains. The megasporangium membrane is distinct, but structureless, and no traces of endosperm or archegonia were found.

The second part of the paper will be devoted to a discussion of the general bearing of the investigation and the probable affinities of the seed. It is to be regretted that in such a paper the citation of literature is not more specific. Reference merely to volume numbers puts needless work upon those who wish to consult the literature.—J. M. C.

**Dorsiventrality in mosses.**—NEMEC has continued his study of dorsiventrality induced by light, finding additional species<sup>14</sup> in which this character can be altered. The tenacity with which dorsiventrality is retained under changed illumination differs widely. Thus in *Anomodon villosus* every flank in turn, as more strongly illuminated, may become the dorsal side. In *Neckera* and *Homalia*, on the contrary, only the dorsal and ventral sides could be made to exchange relations. A geotropic species are usually strongly diaheliotropic (but may become positively clinoheliotropic), turning the dorsal side toward light. Geotropic species are hindered from acquiring a diaheliotropic position, especially if they must curve nearly to the horizontal. The species whose dorsiventrality is not reversible sometimes do not attain completely the diaheliotropic position if much bending is required. Such incomplete reactions are still more common among leafy Jungermanniaceae.

In a later paper on the same lines<sup>15</sup> NEMEC deals especially with *Fissidens* spp., and *Dicranum scoparium*. In *Fissidens* the dorsiventrality (morphological as well as physiological) can be abrogated and recalled at will. The stems are also diaheliotropic and geotropic. In *Dicranum scoparium* the lateral curvature of the leaves is a heliotropic response which is induced very early and persists for a considerable time even in darkness, but finally disappears, returning again with one-sided illumination. The complicated interrelations of geotropism and heliotropic dorsiventrality are worked out in detail.—C. R. B.

**Galvanotropism of roots.**—A paper on this subject, published in September 1904 by Dr. AMOS PLOWMAN,<sup>16</sup> has come to our attention since writing the notice of the papers by SCHELLENBERG and GASSNER.<sup>17</sup> The methods used and the conclusions reached are anticipatory, in large measure, of those of GASSNER. PLOWMAN used carbon electrodes and states the strength of currents used in terms of current density, those mentioned lying mostly between 0.1 and 1.5 milli-

<sup>14</sup> NEMEC, B., Die Induktion der Dorsiventralität bei einigen Moosen. II. Bull. Int. Acad. Sci. Boheme 11: (1-7). 1906.

<sup>15</sup> NEMEC, B., Die Symmetrieverhältnisse und Wachstumsrichtungen einiger Laubmoose. Jahrb. Wiss. 43:501-579. figs. 33. 1906.

<sup>16</sup> PLOWMAN, A., Electrotropism of roots. Am. Jour. Sci. IV. 18:228-236. pls. 9, 10. 1904. Also earlier reports *ibid.* 14:131. 1902, and 15: 94-104. 1903.

<sup>17</sup> BOT. GAZETTE 43:218. 1907.

ampères per cm<sup>2</sup>.sec. He finds the positive curvature constant and shows clearly that it is due to injury of the cells on the positive side. He does not find the negative curvature constant in any case, but observes that the positive curvature (which he calls normal) may be produced, even when the negative appears, by varying the strength and time of action of the current. This GASSNER confirmed, but he paid more attention to the negative curvature as the initial effect of very weak currents, and lays stress on it in identifying galvanotropism with traumatropism as an irritable phenomenon.

PLOWMAN ascribes the injury causing positive curvatures to positive electrons bombarding the side of root turned toward the anode (wherefore he prefers *electrotropism*). He specifically denies the influence of products of electrolysis. Negative electrons he finds innocuous and perhaps even stimulative. Thus his facts are mainly corroborated by GASSNER, but the two interpretations are quite different. GASSNER also seems to have entirely overlooked PLOWMAN's papers.—C. R. B.

**Morphogenesis in Ceratophyllum.**—A recent publication by the Carnegie Institution,<sup>18</sup> under the authorship of Dr. PEARL and assistants, proves to be a contribution of unusual merit because of the masterly way in which the biometrical method has been applied to one of the most important but neglected problems of biology. Two laws of growth in *Ceratophyllum* have been formulated on the basis of convincing evidence. The first law is: "The mean number of leaves per whorl increases with each successive whorl, and in such a way that not only does the absolute increment in each leaf-number diminish, but also the rate of increase diminishes as the ordinal number of the whorl, measured from a fixed point, increases." The second law is: "As whorls are successively produced by a growing bud, they are formed with ever-increasing constancy to their type, the ultimate limit towards which the process is tending being absolute constancy." The significance of these laws and the field for physiological study suggested by them is far too great to be elaborated here and reference to the original is urged. The paper shows a clear scientific discernment and is a model of composition. It may safely be said that the Carnegie Institution has not wasted any money on this minor grant. The value of such a contribution as compared with its insignificant cost goes far toward compensating for the cost of other small grants which have proved less fruitful.—RAYMOND H. POND.

**Relation of saprophytic fungi to the substratum.**—Two papers have appeared dealing with the relation of some saprophytic fungi to their substratum. The first of these, by KRATZ,<sup>19</sup> deals with pyrenomycetes growing on dead stems of herbaceous plants and on leaves. It consists mostly of brief categorical descrip-

<sup>18</sup> PEARL, RAYMOND, with the assistance of OLIVE M. PEPPER and FLORENCE J. HAGLE, Variation and differentiation in *Ceratophyllum*. Pub. Carnegie Institution of Washington no. —. pp. 136. 1907.

<sup>19</sup> KRANTZ, CARL VON, Ueber die Beziehungen der Mycelien einiger saprophytischer Pyrenomyceten zu ihrem Substrat. *Hedwigia* 46:1-24. figs. 8. 1906.

tions of the fruit-bodies and mycelia of the fungi considered. The author attempts to distinguish form-types of perithecia according to their position on or their relation to the elements of the stem; for example, the perithecia originating under the sclerenchyma sheath are flattened, while those originating under the epidermis are round. That pressure from any cause will influence the form of perithecia is self-evident and of little importance. One general fact to be deduced from the paper is that the sclerenchyma tissue is not penetrated by the fungous hyphae.

The second paper, by DUYSEN,<sup>20</sup> deals with wood-inhabiting discomycetes. In this it is shown that the mycelium of many of these fungi, as *Helotium*, *Coryne*, *Mollisia*, and others, penetrates the cells only by the pits or by cracks and fissures caused by other agencies. Enzyme action here is apparently of less importance in aiding the hyphae to penetrate cell walls. In others, like *Propolis* and *Schizoxylon*, more or less of the wood is actually destroyed by enzymosis, mostly in the vicinity of the fruit-body. The sclerenchymatous tissue is not injured.—H. HASSELBRING.

A study in endemism.—Mount Ritigala is an isolated mountain in north-central Ceylon, within 100 feet of whose summit there occurs a remarkable flora, covering only a few acres. In this small space there exist species that belong to the flora of the wetter districts of Ceylon, a regular oasis of vegetation distinct from the dry country surrounding it. WILLIS<sup>21</sup> has studied the possibilities of the origin of this isolated flora and its endemic forms. The number of species enumerated is 144, but 41 of these are eliminated as occurring also in the dry zone and so are not compelled to travel 40 miles to reach the summit. The 103 remaining species all belong to the moist region of Ceylon, at least 40 miles away. Of these species, 24 can be carried by birds, 49 by the wind, and the distribution methods of the remaining 30 are doubtful. After various analyses of the statistics, the conclusion is reached "that endemism, other things being equal, goes in general with difficulty of distribution, and with rare arrival in one spot." For example, the bird-carried forms arrive more regularly than the others, and there is one endemic form among the 24 species; the wind-carried forms arrive less regularly, and there are three endemic forms among the 49 species; while the plants whose method of distribution is called "doubtful" arrive most rarely, and there are 8 endemic forms out of 24 species, 6 of the 30 species being rejected for a special reason.—J. M. C.

Motor mechanism of *Berberis* stamens.—JUEL<sup>22</sup> has made some interesting observations and experiments on the staminal filaments of *Berberis* and *Cen-*

<sup>20</sup> DUYSEN, FRANZ VON, Ueber die Beziehungen der Mycelien einiger, hauptsächlich holzbewohnender, Discomyceten zu ihrem Substrat. *Hedwigia* 46:25-56: figs. 7. 1906.

<sup>21</sup> WILLIS, J. C., The flora of Ritigala, an isolated mountain in the north-central province of Ceylon; a study in endemism. *Annals Roy. Bot. Gard. Peradeniya* 3:271-302. 1906.

<sup>22</sup> JUEL, H. O., Einige Beobachtungen an reizbaren Staubfäden. *Bot. Stud.* tillägn. F. R. KJELLMAN I-20. l. 1. Upsala. 1906.

taurea, though he has not approached nearer a final explanation of the phenomenon than others before him. A cross-section of the filament shows a dorsal region of collenchyma-like tissue with almost imperceptible intercellular spaces. The ventral region is occupied with loose parenchyma. The walls of the collenchyma-like tissue are characterized by considerable contractility in longitudinal direction. The papillose epidermis of the ventral side is practically impermeable, but if carefully removed and the exposed tissue submerged in solutions of varying osmotic strength it appears that a curvature of the filament results from the plasmolysis and consequent contraction of the collenchyma-like tissue. The author's explanation agrees with that of PFEFFER, namely, filtration under turgor pressure, and whatever causes the escape of water from the cell permits contraction of the cell walls and results in curvature. The author thinks that the papillose epidermis of the ventral side cooperates with the collenchyma-like tissue in contraction. Why the contact stimulus should cause filtration under pressure is no more accounted for than before. The filaments of *Centaurea* will endure surprising transpiration without loss of contractile capacity.—RAYMOND H. POND.

The fertile spike of Ophioglossaceae.—CAMPBELL<sup>23</sup> has studied the morphology of the sporophyll and the distribution of the vascular bundles in the three genera of Ophioglossaceae, abundant material having been obtained during his recent visit to the oriental tropics. A study of the development of the sporophyll suggested that the "fertile spike" is the terminal structure rather than an adaxial branch. This view was confirmed by the distribution of the vascular bundles of the leaf, those supplying the spike evidently not being given off from the main bundles of the petiole as branches, but being a continuation of the main bundles. From this point of view, the fertile spike terminates the main axis, which bears the foliage blade as a lateral member. This accords with the author's previously expressed view that *Ophioglossum* is perhaps the most primitive of the pteridophytes, the "fertile spike" being most naturally connected with such a structure as the sporogonium of *Anthoceros*. "This hypothesis assumes that, by the development of a root from the lower part of the sporophyte and a complete septation of the sporogenous tissue of the sporogonium, so that something resembling the spike of an *Ophioglossum* resulted, there would be formed a plant not very unlike *O. simplex*."—J. M. C.

A cotton fungus.—According to a preliminary paper by BALL,<sup>24</sup> the "sore-shin fungus" which occurs on cotton and other plants in the United States is also very common on cotton seedlings in all parts of Egypt. The fungus is said to be responsible for most of the resowing which is necessary in Egyptian cotton fields. The damage, which is not great, is caused partly by rotting of the seed-

<sup>23</sup> CAMPBELL, D. H., Studies on the Ophioglossaceae. Amer. Nat. 41:139-159. figs. 17. 1907.

<sup>24</sup> BALL, W. L., The physiology of a simple parasite. Reprint, Jahrb. Khedivial Soc. pp. 25. pls. 6-7. 1906.

ling in the soil and partly by damping off. A large proportion of the plants which are attacked after breaking through the ground recover by subsequent healing of the injured shins. After showing that the fungus is dependent upon oxygen for growth, the writer states his belief that the fungus attacks the plants at the surface of the ground because here only there exists a zone with sufficient oxygen and yet not too dry for the growth.

It may be mentioned that the sore-shin fungus of the United States is believed by DUGGAR and STEWART<sup>25</sup> to be *Rhizoctonia*. BALL, however, failed to find the typical sclerotia of this genus in the Egyptian form, so that it is possible that the two forms are not identical, unless the sclerotia have been overlooked.—H. HASSELBRING.

**Electroculture.**—GASSNER, before he was called to the University of Montevideo, had begun some investigations on the effect of electric currents, constant and alternating, which, though incomplete, had some interesting results.<sup>26</sup> The constant current in general proved injurious (*cj.* his results and PLOWMAN'S on galvanotropism noted in this journal). The alternating current, when the alternations were rapid enough, was not injurious because it did not act at all; it was not found advantageous, as LÖWENHERZ reports.<sup>27</sup> It does kill grubs and worms in the soil when not hurtful to plants and may thus be useful practically. With induced currents from a frictional machine, potted barley grew better when electrified for 13–14 hours daily from needle-points above the plants, which accords with the earlier results of LEMSTRÖM (1890) and CHODAT (1892). One notable fact was the threefold evaporation from the electrified plants, as compared with the controls. Even greater differences, 6:1, appeared when porcelain dishes filled with water were substituted for the pots.—C. R. B.

**Palisade cells.**—RAUNKIÆR decides in favor of STAHL'S view, that palisade cells owe their form to light, as against ARESCHOUG'S that transpiration is the determining cause, from his studies upon the palisade of *Scirpus lacustris*.<sup>28</sup> The leaves of plants growing somewhat apart from the clump show equally well-developed palisade cells in the leaf above water and to a depth of 20<sup>cm</sup>. From this point to a depth of 50<sup>cm</sup> the length of the palisade cells gradually diminishes. In similar plants growing crowded and therefore shaded, no palisade cells are distinguishable in the submerged parts, nor in the aerial region up to a height of 30–40<sup>cm</sup>, where they commence to appear. In this case there is clear evidence that the external factor, light, calls forth this differentiation. In other

<sup>25</sup> The sterile fungus *Rhizoctonia*. Cornell Univ. Agr. Exp. Sta. and N. Y. Agr. Exp. Sta. Bull. 186. 1901.

<sup>26</sup> GASSNER, G., Zur Frage der Elektrokultur. Ber. Deutsch. Bot. Gesells. 25:26–38. 1907.

<sup>27</sup> LÖWENHERZ, Versuche über Elektrokultur. Zeit. Pfl.-krankh. 15:137 ff. 1905.

<sup>28</sup> RAUNKIÆR, C., Nogle Iagttagelser og Forsøg over Aarsagerne til Palissadecellernes Form og Stilling. (Sur les causes qui déterminent la forme et l'orientation des cellules palissades.) Bot. Tidsskrift 27:293–311. 1906.

plants it may have become hereditary, i. e., due to internal causes. RAUNKIAER finds in certain plants also that direction of light determines the orientation of the palisade cells; in the great majority, however, their orientation is established independent of this factor.—C. R. B.

**Inheritance and gynodioecism.**—CORRENS<sup>29</sup> makes a third report on the inheritance of sex in gynodioecious plants, the species dealt with being *Satureia hortensis* and *Silene inflata*, previously reported on, and in addition, *Silene dichotoma* and *Plantago lanceolata*. All of these species confirm the two laws previously derived by the author, namely, that in gynodioecious plants each sex-form produces gametes which have the tendency to produce the same sex-form, and that the phylogenetically newer unisexual form dominates more or less completely the older bisexual form. There seems to be a third class in the case of *Plantago lanceolata*, characterized by an intermediate condition. These are described as gynomonoecious or as stunted bisexual. The offspring of these contained 28–32 per cent. ♀, as compared with 0–3 per cent. among the offspring of normal bisexual parents, and 71–96 per cent. among the progeny of fully pistillate individuals.—G. H. SHULL.

**Commercial forage seeds.**—The adulteration of commercial forage seeds has become a question of great importance in the western agricultural regions, and reliable, easily applied distinctions between the adulterants and the genuine seed are being sought for energetically at the Agricultural Experiment Stations. Two recent bulletins from Kansas deal with two of the most important forage crops, namely alfalfa<sup>30</sup> and blue grass.<sup>31</sup> To recognize without fail the commercial seed of alfalfa is to protect one of the largest financial interests of the region. In the case of the distinction between the grains of *Poa pratensis* and *Poa compressa*, it is interesting to note that these observers have discovered what seems to be an unfailing character in the toothing of the palet, which agrostologists have overlooked.—J. M. C.

**Progress of accretion during growth.**—As stated by HEDLUND,<sup>32</sup> it is important to ascertain the amount or rather the rate of accretion correlated with internal processes of the protoplasm during growth. This author has made a quantitative study which is certainly a step in advance. He finds that a single cell (alga) grows more rapidly when free than when in contact with a neighbor. Growth declines for a period before division and remains slow while the protoplasm is

<sup>29</sup> CORRENS, C., Die Vererbung der Geschlechtsformen bei den gynodiöcischen Pflanzen. Ber. Deutsch. Bot. Gesells. 24:459–474. 1906.

<sup>30</sup> ROBERTS, H. F., and FREEMAN, G. F., Alfalfa seed; its adulterants, substitutes, and impurities, and their detection. Kans. Agric. Coll. Bull. 133. 1906.

<sup>31</sup> ———, Commercial seeds of brome grass and of English and Kentucky blue grasses; adulterants and substitutes and their detection. Kans. Agric. Coll. Bull. 141. 1907.

<sup>32</sup> HEDLUND, T., Ueber den Zuwachsverlauf bei kugeligen Algen während des Wachstums. Bot. Stud. tillägn. F. R. KJELLMAN 35–54. pls. 4. Upsala, 1906.

engaged in division. The figures support what the author regards as a general law, that the rate of accretion varies inversely with the size of the organism. There is nothing particularly new about that, but there is value in abundant data.—RAYMOND H. POND.

**Mechanics of plants.**—LORCH describes the arrangement of mechanical tissues in a number of mosses and the warping effects produced thereby in absorbing or losing water.<sup>33</sup> His observations upon the circinate inrolling of *Leptodon Smithii* and some allies, and the behavior of leaves possessing mechanical tissues in ribs and borders, as the Polytrichaceae, are interesting, but develop nothing specially novel. The same may be said of the study of the mechanical system of the hyaline cells in Sphagnum leaves.<sup>34</sup>—C. R. B.

**Respiration.**—A rather startling announcement is that by STOKLASA<sup>35</sup> and his assistants that in coal and lignite they find an enzyme, peroxidase; and by comparative experiments on sterilized and non-sterilized coals, following the methods of PALLADIN and his pupils, they recognize the excretion of CO<sub>2</sub>, as dependent partly upon autoxidation and partly upon enzymic action. The excretion of methane and hydrogen is due to the peroxidase.—C. R. B.

**Secretions of enzymes.**—PANTANELLI has continued his study of this topic, of which he presents a detailed account.<sup>36</sup> Though he has recorded the effect of many substances upon the formation and action of invertase in *Mucor*, he has not been able to throw much light upon the deeper problem of the method of secretion.—C. R. B.

**Physiology of movement.**—The *New Phytologist* is printing an interesting series of lectures on this subject by Mr. FRANCIS DARWIN, beginning in the number for November 1906.<sup>37</sup>—C. R. B.

<sup>33</sup> LORCH, Wm., Einige Bewegungs- und Schrumpfungerscheinungen an den Achsen und Blättern mehrerer Laubmoose als Folge des Verlustes von Wasser. *Flora* 97:76-95. figs. 20. 1907.

<sup>34</sup> ———, Das mechanische System der Blätter, insbesondere der Stämmchenblätter von Sphagnum. *Idem* 97:196-206. figs. II.

<sup>35</sup> STOKLASA, J., A. ERNST and K. CHOCENSKÝ, Ueber die anaërobe Atmung der Samenpflanzen und die Isolierung der Atmungsenzyme. II. Ber. Deutsch. Bot. Gesells. 25:38-42. 1907.

<sup>36</sup> PANTANELLI, E., Meccanismo di secrezione degli enzimi. *Annali di Bot.* 5:229-272, 355-416, 1906.

<sup>37</sup> I. Associated stimuli. Nov. 1906. II. On some questions of nomenclature and method. Dec. 1906. III. The analysis of geotropism. Jan. 1907. IV. The localisation of perception. Feb. 1907.

## NEWS

DR. EDWARD C. JEFFREY, Harvard University, has been promoted to a full professorship of plant morphology.

DR. K. M. WIEGAND, Cornell University, has been appointed associate professor of botany in Wellesley College.

PROFESSOR FRANCIS E. LLOYD, of the Desert Laboratory, is announced for a course in botany during next July at the summer school of Harvard University.

THE MACMILLAN COMPANY has announced the publication of a "Practical text-book of plant pathology" by D. T. MACDOUGAL, F. S. EARLE, and H. H. RICHARDS.

DR. K. MIYAKE, Doshisha College, Japan, has been selected to fill the position of Professor IKENO at the University of Tokyo during the two years' absence of the latter.

THE LAKE LABORATORY of the Ohio State University is located on Cedar Point, near Sandusky, and will hold the session of 1907 from June 24 to August 2. MALCOLM E. STICKNEY, Denison University, will be in charge of the botany.

DR. MEL T. COOK has been appointed plant pathologist to the Delaware Agricultural Experiment Station, under the recent Adams Act. He is also to act as professor of botany in Delaware College. His future address will be Newark, Del.

DR. JACOB SCHNECK, Mount Carmel, Ill., died December 18, 1906. Dr. Schneck was an old and well-known student of the flora of his region, and his name is a very familiar one to systematists. A number of his contributions appear in the earlier volumes of the BOTANICAL GAZETTE.

THE SEVENTH ANNUAL SEASON of the Minnesota Seaside Station, on the Straits of Fuca, Vancouver Island, is announced for the six weeks beginning about July 6. Full information may be obtained from Assistant Professor JOSEPHINE E. TILDEN, University of Minnesota, Minneapolis.

PROFESSOR W. A. KELLERMAN, Ohio State University, has returned from an absence of three months in Guatemala, where he has been collecting parasitic fungi. He has planned a "Peripatetic School of Tropical Botany" for a limited number of students, the first session to begin December 20, 1907.

SIR THOMAS HANBURY, the owner of the well-known Botanical Garden of La Mortola, Ventimiglia, Italy, died March 9, after an illness of a few weeks. He would have completed his seventy-fifth year on June 21. His death will be deeply mourned by many botanists who have visited his beautiful garden, and who have known him personally.

THE MARINE BIOLOGICAL Laboratory, Woods Hole, Mass., will begin its twentieth session on June 1, 1907. Dr. GEORGE T. MOORE will be in charge of the botany, and associated with him there will be GEORGE R. LYMAN, Dartmouth College, and IVEY FOREMAN LEWIS, Johns Hopkins University. DR. BRADLEY M. DAVIS has resigned.

DR. OTIS W. CALDWELL, State Normal School, Charleston, Ill., has been added to the botanical staff of the University of Chicago, as associate professor of botany in the College of Education, and DR. CHARLES J. CHAMBERLAIN and DR. HENRY C. COWLES have been promoted from instructorships to assistant professorships in the department of botany.

AT THE NEW YORK MEETING of the American Association a silver loving-cup was presented to Professor W. F. GANONG. The presentation was made by Professor G. F. ATKINSON on the part of the former members of the Society for Plant Morphology and Physiology, as a token of appreciation of Professor GANONG'S very efficient services during ten years as executive officer of that society.

THE BIOLOGICAL LABORATORY of the Brooklyn Institute, which conducts summer work at Cold Spring Harbor, has announced that its next annual session will be held during July and August, 1907. The botanical staff will be as follows: Dr. D. S. JOHNSON, Cryptogamic Botany; Dr. E. N. TRANSEAU, Plant Ecology; Mr. H. H. YORK, Associate in Botany; Mr. W. S. COOPER, Assistant in Ecology.

PROFESSOR B. E. FERNOW, State College, Pennsylvania, has been appointed dean of the Faculty of Forestry at the University of Toronto. According to *Science*, "it is proposed to organize the most complete system of forestry education on this continent, the university undergoing now a reorganization and broadening out its policy generally. It is expected that a large forest reservation will be set aside for the use of the school, and university extension work of the broadest kind is contemplated."

DR. OTIS W. CALDWELL, State Normal School, Charleston, Ill., has made several visits to Cuba in search of Microcycas, reported as being endemic on the island and unknown in collections in any reliable way. The last trip was made during February and early March, and resulted in complete success. He has secured material for a proper morphological study of the genus, and his results will be an important contribution to our accumulating knowledge of the morphology of the cycads.

AN ASSOCIATION of twelve botanists has undertaken the direction of the *Plant World*, beginning with 1907. The scope of the journal will be enlarged by including more notes and news, accounts of explorations, illustrations of experiments, discussions of evolution and plant-breeding, etc. The subscription price is to remain \$1 a year, and F. E. LLOYD continues to be the managing editor. The association includes the following botanists: J. C. ARTHUR, Miss M. M. BRACK-

ETT, N. L. BRITTON, W. A. CANNON, W. F. GANONG, D. S. JOHNSON, B. E. LIVINGSTON, F. E. LLOYD, D. T. MACDOUGAL, W. B. MACCALLUM, V. M. SPALDING, and J. J. THORNBERRY.

THE THIRD annual meeting of the Botanists of the Central States was held at Madison, Wis., March 28-30, in connection with the annual meeting of the Central Section of the American Society of Naturalists. About thirty were in attendance and the programme of papers continued until noon of the third day. The dominant note of the programme was cytology. Thursday afternoon was devoted to demonstrations of microscopic material, apparatus, cultures, etc., of which there was an instructive display. A smoker and the annual dinner of the naturalists gave opportunity for social acquaintance. President NEWCOMBE'S address, "A need of botanical science in America," was devoted to showing the need of additional means for the publication of research. The Botanical Society of America was requested by resolution to take up the matter. Professor T. H. MACBRIDE, University of Iowa, was elected president.

# BOTANICAL GAZETTE

JUNE 1907

## THE INFLUENCE OF TENSION ON THE FORMATION OF MECHANICAL TISSUE IN PLANTS\*

RUFUS PERCIVAL HIBBARD

The radical change from a theory of self-regulatory development of mechanical tissue in plants subjected to tension by pull, to one implying no reaction whatever, necessitates detailed evidence before the latter can be accepted. The following investigation was directed to a more complete knowledge of the reactions of a plant to tension, to determine if possible what influence this exerts on the formation of mechanical tissue. Heretofore the root system has been entirely ignored, but this together with the stem has been included in the work here recorded.

### I. Historical

For some time previous to 1891, physiologists had generally assumed that a plant reacted to a gradually increasing strain by a development of its mechanical tissue. HEGLER (PFEFFER '91) in this year subjected several stems and leaves to a longitudinal pull. In response to this method of experimentation, the plant, according to HEGLER, withstood greater strain than one grown under normal conditions. For example, a seedling of *Helianthus annuus* whose original breaking strength was 160<sup>gm</sup> had a breaking strength of 250<sup>gm</sup> after two days under the influence of a pull of 150<sup>gm</sup>. The petioles of *Helleborus niger*, which at first could withstand a weight of only 400<sup>gm</sup>, after five days, during which the weights had been gradually increased, held without breaking 3.5<sup>kg</sup>, while those under normal conditions gained but little strength in the same time. Marked differences were

\* Contribution 92 from the Botanical Laboratory of the University of Michigan.

apparent, not only in the tensile strength, but also in anatomical structure. On examination of various sections, HEGLER noticed (1) a numerical increase in the cells of the collenchyma; (2) an increased thickness of the walls of the collenchyma, sclerenchyma, and bast; and (3) the production of entirely new tissues. These observations led him to the conclusion that a plant responds to a gradually increasing strain by a development of mechanical tissue. It is indeed unfortunate that only a preliminary report of HEGLER's work can be had. The influence that his conclusions exerted has been widely felt, and the principles laid down were widely accepted.

RICHTER ('94) concluded that when the stems of Chara were pulled longitudinally, there was an increase of strength. Thus, in his opinion, he confirmed the observations and results of HEGLER. But we may dispose of this interpretation in the words of BALL (:04), who says: "Die Resultate RICHTERS sind etwas zweifelhaft, da er keinen Vergleich zwischen belasteten Chara-Pflanzen und unbelasteten von demselben Alter und derselber Grösse gegeben hat."

VON DERSCHAU ('94) came to the conclusion that a gradually increasing pull without contact on certain twining petioles raises the breaking strength, increases the development of mechanical tissue, and calls forth the development of new tissue. His first statement is borne out by experiments which show that even after twenty-four hours the plant part under strain could withstand a weight which at first would have broken it. From a study of sections of the various petioles, he found that marked anatomical differences occurred. There was a numerical increase in almost all kinds of cells. The bast thickened in all the petioles examined except those of *Solanum jasminoides*, and the ring of vascular bundles was made complete in all but *Solanum*, where it was half-moon shape, as in the normal petioles.

Concerning tendrils no experiments have been recorded to determine the effect of pull alone on the free basal portion. PENHALLOW ('86, p. 49) noted that tendrils that had secured attachment were larger, stronger, and more rigid, from which it would appear probable that contact produces a more or less marked effect in accelerating or at least in increasing the strength of parts. This view gains strong confirmation from similar conditions in *Ampelopsis*.

WORGITZKY ('87), in his "Vergleichende Anatomie der Ranken," gives the results of weighting experiments, and comes to the same conclusions as von DERSCHAU. He found, by comparing weighted tendrils of *Passiflora quadrangularis* which had secured attachment with weighted tendrils which had not, that the tensile strength of the former had increased about twice as much. With tendrils of *Cucurbita Pepo* the resistance to strain was increased thirteen fold. This he believed was not the result of pull alone, but one of pull and contact combined.

NEWCOMBE ('95, p. 446), speaking of the reaction of tendrils to contact, states "that the first strengthening tissue is here laid down as a response to contact; its increase is the regulatory response of the plant to the strain that it feels."

MACDOUGAL ('96, pp. 377, 378) believes that contact stimuli are not transmitted beyond 2 or 3<sup>mm</sup> and, as the thickening of the tendril always takes place after contact, we are left to conclude that it is due to the pull or traction exerted by the weight of the stem supported by the tendril.

PEIRCE (:04, p. 241) believes also that the strengthening of the free basal portion is not due to contact, but to the pull exerted in bringing the stem nearer the support.

FITTING (:03, p. 476) has lately shown that contact stimuli are transmitted for some distance, and for that and other reasons their effect on the basal portion of the tendrils cannot be excluded. It thus becomes a question how much of the increase of strength is due to pull and how much to contact.

VÖCHTING (:02) investigated the influence of pull on sunflowers and cabbages that had been prevented from flowering by means of decapitation, and found that no new tissue had been formed and that no increase of mechanical tissue had occurred as a result of the pull.

WIEDERSHEIM (:03) finds that the expected development of new tissue and the thickening of the wood and bast fibers in weighted pendent branches of *Fraxinus*, *Fagus*, *Sorbus*, and *Ulmus* does not occur. On the other hand, in *Corylus* he observed an increase in the number of bast fibers. This he attributes to a self-regulatory development.

BALL (:04) repeated the work of HEGLER. As a result of numerous

experiments to determine exact conditions, he has been able to show that in general the breaking strength of a plant stem does not increase in response to a gradually increasing pull, and that no increase of breaking strength follows as a result of pull on a stem growing in a horizontal plane. Detailed examinations of microtomic and free-hand sections, stained and unstained, show that there is, as a result of pull, no increase in the thickness of the walls, nor a numerical increase in the cells of the various tissues. No new tissues are produced as a result of the strain. A number of other authors, KÜSTER (p. 173), PFEFFER (p. 101, p. 148), and VÖCHTING (p. 282), have also failed to detect or observe any production of new tissues.

The factor of correlation has yet to be briefly mentioned. It is generally admitted by writers who have investigated the subject that a very high degree of correlation is manifest between the various organs of the plant. GOEBEL writes in his *Organography of Plants* (p. 206) "that careful research demonstrates the existence of reciprocity between parts of the plant body. . . . The size and construction of one organ are frequently determined by those of another."

KLEIN ('86) showed that the bundles were more centrally located in the fruit stalk than in the petiole, and attributed this arrangement to the necessity for a greater mechanical strength, as well as for a more abundant supply of building material.

DENNERT ('87) on comparing the anatomical structure of the fruit stalk, before and after ripening of the fruit, found an increase in the development of mechanical tissue. This was apparent in the greater increase in the xylem and in the thickness of walls of the wood fibers.

REICHE ('87) corroborates the work of the earlier investigators and shows in many additional plants that changes in the flower stalk during its transformation to a fruit stalk go hand in hand with the development of the fruit.

PIETERS ('96) showed that although one-year-old fruit-bearing shoots of the apple and the pear had a smaller xylem cylinder in proportion to their diameters than the vegetative shoots of the same age, they were well supplied with supplementary mechanical tissues which was distributed at those points where it was most needed. In the case of the peach and the plum, the woody cylinder was larger in the fruit-bearing shoot than in the vegetative shoot. Fruit-bearing also

exerted an influence on the lignification of the cell walls of many of the tissues in the stalk. In the apple and the pear there was an abundance of well-lignified sclerenchyma and hard bast, which occurred in the vegetative shoot only sparingly, if at all.

BOODLE (:02) states that the walls of the sieve tubes and companion cells in *Helianthus annuus* become lignified as a result of strain. We also read "that the slight lignification of the parenchymatous parts of the pericycle and medullary rays unites the primary sclerenchyma strands into a more definite mechanical system attached to the strong xylem by the medullary rays." Finally, he says, "this must give greater rigidity, which no doubt is required by the heavy fruiting capitula borne by the plant."

On the other hand, in opposition to this theory, KELLER (:04) finds that pull as such does not call forth a regulatory strengthening of mechanical tissue in fruit stalks. Fruit-bearing in itself does not cause a thickening of parts nor exert an influence on the lignification of the cell walls. Upon orthotropic flower stalks, a strong or light pull in the direction of the long axis exerts no influence on the development of mechanical tissue. Displaced stalks under tension show no self-regulatory thickening, but certain anatomical changes do take place. These changes are not due to tension, but simply to the alteration in the position from orthotropic to plagiotropic, and in turn are directly referred to differences in the degree of strain between the upper and lower sides. These conclusions do not fall in line with the view that has held ground for some time, namely, that the mechanical development of the stalk goes hand in hand with the development of the fruit. Correlative growth, it is said, is no explanation for this phenomenon. This interpretation is yet to be justified. If this be true, our previous ideas must undergo transformation.

As a conclusion of the historical part of this paper I might summarize these views: Tension has no influence on the increase of mechanical tissue in any stems examined, in any petioles, nor in branches, except in *Corylus avellana*. It has some effect on twining petioles in that there occurs a thickening and lignification of certain tissues. The effect on tendrils has not yet been accurately determined. Until further data are gathered to show the contrary,

compensatory regulation or correlative growth cannot be considered as an unimportant factor.

## II. Methods

The seedlings and plants examined in my work were *Helianthus annuus*, *Phaseolus multiflorus*, *Ricinus communis*, *Brassica oleracea*, *Coleus tricolor*, *Fuchsia speciosa*, and *Vinca major*. The stems compared were selected with reference to similarity in size and vigor. All tension and normal plants were grown under similar conditions.

When collected, the stems were numbered, cut into suitable lengths, and put into 50 per cent. alcohol for further examination. To study the stems, freehand cross-sections were made and the tissues measured by several methods. Further description of the different methods will be given under the proper headings.

## III. Results of experimentation

### A. INFLUENCE OF TENSION ON STEMS

A number of seedlings were subjected to tension in the following manner: Strong, light twine was fastened about the stem, which had previously been wound with cotton flannel to eliminate any injury due to the cord. The twine was then run over a lightly rolling pulley, hanging from a support directly above, and the weights attached to the free end of the cord. The twine was so fastened that very little injury, if any, was caused. Two loops were made from short pieces of twine, each about 30<sup>cm</sup> in length. These were noosed about the stem, from opposite sides one within the other, so that four circles of twine surrounded the stem over the cloth. The ends of the loops were then connected by a short piece of twine. To this was fastened, in an adjustable manner, the cord running over the pulley. In this way, if one loop should happen to be longer than the other, an adjustment took place, so that the strain on each loop was the same, in consequence of which the stem felt the pull in the direction of its longitudinal axis.

When the seedlings were young, the unfolding leaves and growing tips might be somewhat hindered in their growth by the interference of the loops of twine. To prevent this, a strip of light whitewood, about 10<sup>cm</sup> long, 3<sup>mm</sup> wide, and not more than 1.5<sup>mm</sup> thick, was so placed between the loops as to form a diamond-shaped figure, such as

has been described by BALL (p. 309). When the plants had reached a more advanced stage of development, this device was unnecessary, and was therefore discarded. To guard further against any possible injury that the cord might produce in the stem when heavy weights were used, the cord was frequently changed to a place above or below the original attachment.

The seeds were germinated in 20<sup>cm</sup> pots containing garden loam and clay. The loam and clay were thoroughly mixed in the proportion of one of garden loam to three of clay. This gave the earth a firm cohesion, and for this reason few seedlings were uprooted. When necessary, a plaster of Paris cap was cast over the earth to the edge of the pot and securely fastened with cord. The plaster was kept from the stems by means of surrounding paper cylinders. When the seedlings had reached a height of 8 to 10<sup>cm</sup>, the experiments were usually begun, although in some cases they were allowed to grow stronger and taller before being subjected to tension.

There are two ways to determine whether or not tension has increased mechanical tissue. First, by a measurement of the breaking strength, and second, by the observation of the size of the various tissues and the thickness of the cell walls. The first method has been followed admirably on various stems by BALL. Beyond a few experiments performed by the writer, the need of further observations by this method seems quite unnecessary.

To determine by microscopic means the differences between tension and normal plants in the mechanical tissues, several methods were used. One method was to determine the thickness of tissues and walls by an ocular micrometer. The areas of the woody cylinders were computed by making camera-lucida drawings of the xylem, and the areas of these irregular drawings were determined by means of a polar planimeter. A third method was devised of making camera drawings of the woody cylinder on Bristol board, which were then carefully cut out and weighed on a chemical balance. A fourth method consisted in comparing the phloem bundles of the tension and normal plants. The number of bast fibers in cross-section of a tension plant was compared with that in the bundles of the normal plant. The number of bundles in one was directly compared with the number in the other. A fifth method consisted in comparing

sections of the tension-plant stem above and below the place of attachment of the cord, to note any differences in anatomical structure in the two regions.

*Helianthus annuus* (greenhouse cultures)

Ten plants of *Helianthus annuus* that had grown in the greenhouse for a month were weighted on November 4 with 50<sup>gm</sup>. On the third day following, the weight was increased to 100<sup>gm</sup>, and on the tenth to 200<sup>gm</sup>. At this time the attachments were changed to a place above the original one. This, as has been mentioned above, was for the purpose of guarding against injury due to the cord. Additional weights were added at various times, until in this set of experiments the final amount reached 600<sup>gm</sup> on November 23.

At this time the weights were removed from five plants and the stems placed in 50 per cent. alcohol. Five plants that had grown under similar conditions, except that they had not been subjected to tension, were also gathered to serve as controls. These controls had been chosen at the beginning of the experiment and were closely watched throughout the time of experimentation in order that all conditions other than tension should be the same.

Cross-sections of the normal stem of *Helianthus annuus* of this age, at a height of 7<sup>cm</sup> from the ground, do not show a completely closed xylem ring. The bundles are separate and are of various sizes. The secondary xylem is beginning to form.

Sections for microscopical study were made at a place 7<sup>cm</sup> above the first lateral root in both tension and normal plants. The place of attachment of the cord on the tension plant was some distance above this, so the sections were within the region of tension. Careful observations by two persons, working independently, did not disclose any appreciable difference between tension and normal plants.

Other sections were then made in the tension stem above and below the attachment. No appreciable differences are detected when sections in the tension region are compared with sections on the same plant out of the region of tension.

As a general result of observations of the first five plants, we may say that tension exerted upon the stem along its longitudinal axis did not call forth a self-regulatory development of mechanical tissue.

The remaining five plants of the ten used in the above experiment were allowed to grow with the constant pull of weights until they had produced flowers. The purpose of this experiment was to note the influence of tension continued for a long period. These plants because of the winter season were rather weak. Cross-sections of these stems at 7<sup>cm</sup> above the ground show a closed xylem ring. The area and weight methods as described above were employed for detecting differences. Even in these examples we find no self-regulatory development of mechanical tissue in response to tension, nor are the results on old stems any different from those on young ones. The final weight carried by each plant was 2300<sup>gm</sup>.

The above experiments on *Helianthus annuus* were repeated with twenty-five other individuals and the same methods for detecting differences were used. Only a few of these plants were allowed to mature, as young plants give just as reliable results and are much more easily handled. The duration of the experiments varied from two to four weeks. The final weights varied from 600<sup>gm</sup> to 2860<sup>m</sup>. The general result was that tension, gradually increased and in the direction of the longitudinal axis, did not induce a self-regulatory development of mechanical tissue in the sunflower stem. In this we agree fully with the results of BALL.

#### *Helianthus annuus* (field cultures)

Seventeen sunflower plants were subjected to the same condition of strain, but to other external conditions which were not so uniform as those in the greenhouse. These experiments were conducted in the open field during the months of July and August 1905.

An examination of the cross-sections of these stems, taken at 1<sup>cm</sup> above the first lateral root, gives the general result that was found to be true in the case of the plants grown in the greenhouse, namely, that an increasing tension along the longitudinal axis does not call forth a self-regulatory development of mechanical tissue in the sunflower stem. By the area method only three tension plants show an increase of mechanical tissue over that of the controls. Two control plants show an increase in mechanical tissue over that of the tension. The others show slight differences that fall within the limit of error. Ten examples from the seventeen are given in Table I.

TABLE I

Plant	Tension	Normal	Percentages (Normal=100)
No. 1 A.	29.87 sq. cm.	31.16 sq. cm.	- 4
No. 2 B.	42.45	42.71	- 6
No. 3 B.	17.42	17.29	8
No. 4 B.	23.74	23.23	2
No. 5 B.	35.62	54.52	- 53
No. 10 B.	33.68	51.81	- 54
No. 9 A.	53.16	31.81	67
No. 5 A.	56.13	36.45	54
No. 8 A.	55.04	49.42	11
No. 1 B.	34.65	32.65	6

In the above table the second column contains the measurements of the relative area of xylem in the tension plant in section; the third column contains similar measurements in the control plants in section; and the fourth column contains percentage differences when the normal is taken as 100.

#### *Ricinus communis* (field cultures)

The same experiments were made upon *Ricinus* stems, which were also cultivated in the open field during the summer months. Seventeen, chosen at random, from the large number that were put under tension, showed after close observations the same general results as previously indicated for *Helianthus*. There is no constant increase in mechanical tissues as a result of a gradually increasing pull. The following table shows results of observations on ten plants.

TABLE II

Plant	Tension	Normal	Percentages (Normal=1)
No. 1 A.	24.20 sq. cm.	17.10 sq. cm.	42
No. 2 A.	59.615	39.87	50
No. 3 A.	26.45	23.915	11
No. 5 A.	52.50	59.9	- 14
No. 9 A.	52.39	56.555	- 8
No. 10 A.	45.00	55.7	- 24
No. 3 B.	30.10	24.6	23
No. 4 B.	35.30	33.9	4
No. 5 B.	15.80	15.40	3
No. 7 B.	50.30	48.70	3

Nos. 1A, 2A, 3A, and 3B show results in favor of tension plants; nos. 5A and 10A show results in favor of normal plants; nos. 9A,

4B, 5B, and 7B are within the limit of error. The remaining seven of the original seventeen were estimated with the aid of the microscope. These in nowise alter the general conclusion.

*Vinca major* (greenhouse cultures)

Twenty-three thrifty young stems of *Vinca major*, some of which were still growing upright, and others that had already bent over and were growing in the usual drooping way, were subjected to tension by means of weights. The young upright stems were placed under tension in the same manner as explained above for *Helianthus annuus*. The pendent stems were placed under tension by hanging the weights upon them without the use of the pulley. Cotton flannel was wound around the stem just above a node. Two loops of twine were then noosed around the stem over the cloth, and drawn tight. From the end of each noose hung equal weights, and thus the stem received the strain along its longitudinal axis.

The duration of the experiments on the upright stems varied from two to four weeks, and the final weights varied from 1250 to 1500<sup>gm</sup>. The duration of the experiments on the pendent stems varied from eleven to twenty-eight days, and the final weights varied from 1000 to 1500<sup>gm</sup>. Normally growing stems for comparison with tension stems were selected with regard to similarity in diameter, length, manner of growth, and vigor. In most cases sections were taken in normal and experimental plants at equal distances from the stem apex.

Observations with a magnification of 100 diameters show that in the majority of plants the whole xylem ring is thicker and composed of thicker-walled cells in the tension plant. The bast is more abundant in the tension plant. Under a higher magnification of 150 diameters, it is noticed that the walls of the bast fibers are thicker in the tension plant.

In all of the twenty-three plant stems under tension, five only do not show an increase in the absolute size of the xylem or in the thickness of the cell walls of bast or xylem. The following tables give the results obtained for a few of the representative plants and their controls. The preponderance of evidence, judging from the number of plants experimented on, shows that an increase in the quantity of xylem amounts to at least 50 per cent. In the table, T indicates

tension plant, and N the control. The areas given are those measured on the drawings.

TABLE III (AREA METHOD)

Plant	Area of whole section	Area of xylem and pith	Area of xylem	Percentages
No. 2A. T.	97.75 sq. cm.	35.62 sq. cm.	15.29 sq. cm.	53
No. 2A. N.	97.62	34.32	10.00	
No. 3 T.	153.49	60.64	20.13	82
No. 3 N.	154.20	61.62	11.03	
No. 2B. T.	129.49	53.75	14.65	- 3
No. 2B. N.	125.10	50.13	15.03	

The last plant (no. 2B. T) is one of three mentioned above. This shows a slight difference in favor of the normal and by so much weakens the general conclusion.

The first four plants of the preceding table were used to test the differences by the weighting method. The two methods show results which are very close, only varying by 2 per cent. to 4 per cent.

TABLE IV (WEIGHT METHOD)

Plant	Weight of drawing of cross-section	Weight of xylem	Percentages Difference
No. 2 T.	1.87 gm	0.28 gm	
No. 2 N.	1.89	0.19	49
No. 3 T.	2.8641	0.382	
No. 3 N.	2.9301	0.215	78

#### *Brassica oleracea* (greenhouse cultures)

Five plants of *Brassica oleracea* were put under tension in a similar way as explained for *Helianthus annuus*. The seeds were planted in 20<sup>cm</sup> pots and placed in the greenhouse. In a little more than a month these had grown sufficiently for experimentation, being about 7<sup>cm</sup> tall. The original weight for each plant was 50<sup>gm</sup>; this was gradually increased until the final weight reached 1000<sup>gm</sup>. The duration of the experiment was two weeks.

Sections were made freehand at places 5<sup>cm</sup> and 1<sup>cm</sup> above the ground in both control and tension plants. An examination of the various anatomical structures failed to show any constant differences between the control and experimental plants. The xylem ring in the

tension plant when compared with the xylem ring in the control plant showed no appreciable difference. The phloem in each had developed to the same degree. Under high power the walls of the cells in the tension plant were seen to be no thicker than the walls of the corresponding cells of the normal plant.

A further study, such as has been made with other stems by determining differences with the aid of area or weight methods, seemed unnecessary. It must be concluded then, as a result of experiments on five plants of *Brassica oleracea*, that tension does not call forth a self-regulatory development of mechanical tissue.

#### *Phaseolus multiflorus* (greenhouse cultures)

Four plants of *Phaseolus multiflorus* were subjected to tension for two weeks. These plants were cultivated in the greenhouse and when a few centimeters high were put under a strain in the same manner as explained for *Helianthus*. Small weights were first attached, and to these were added others until the final weight amounted to 1750<sup>gm</sup>. Freehand sections were made at heights on the stem 5<sup>cm</sup> above the ground. Sections at corresponding places in the control plants were chosen for comparison. Control plants, as in all experiments, were selected with reference to similarity in growth, size, and vigor.

In cross-section the stem of *Phaseolus* shows a very well-differentiated xylem ring, and it is very easy to note differences if they occur, when tension and control plants are compared. In the four plants under experimentation, a careful inspection with the microscope failed to show any appreciable differences in the size of the xylem ring, bundles of phloem, or thickness of walls. For this reason it seemed unnecessary to proceed with the various methods as used in previous experiments. My conclusion, therefore, with reference to the influence of tension on the stem of *Phaseolus multiflorus* can be nothing else than that tension does not induce a self-regulatory development of mechanical tissue.

#### *Conclusion*

From the above seven experiments we conclude that tension has no influence in the formation of tissue in stems of *Helianthus annuus*, *Ricinus communis*, *Brassica oleracea*, and *Phaseolus multiflorus*, but that in *Vinca* it seems to call forth a regulatory development of sup-

porting tissues. This shows itself in an increase in the amount of xylem and in the thickness of the cell walls of the same.

#### B. INFLUENCE OF COMPRESSION ON STEMS

In my experiments I made use of the following plants and seedlings, which were cultivated in the greenhouse: *Helianthus annuus*, *Vinca major*, *Fuchsia speciosa*, and *Coleus tricolor*. The controls were raised in the same place, and received the same care and attention as the plants under experimentation.

A number of the above-named plants and seedlings, young and old, were subjected to compression by various means. The most common means was that of fastening the weights directly upon the stem. Around the stem, some distance above the ground, usually not more than 15<sup>cm</sup>, a strip of cotton flannel was wound. Over this and around the stem from opposite sides were noosed two loops in the manner already described in the tension experiments on *Helianthus*, *Vinca*, etc. Equal weights were attached to each loop, so that the strain was uniform on the opposite sides of the stem. To insure the stem from bending under the weight, bamboo stakes were driven into the ground close to the plant and then fastened to it in two places, one above and one below the place from which the weight hung, but so fastened as not to free the plant from the compression of the weights.

In other cases the compression was narrowly localized. This regional compression was brought about in the following manner: Weights were hung from the sides of the stem as has already been explained. Below the attachment of these weights, some 10<sup>cm</sup> distant, a strong cord was fastened about the stem in the manner as described for the tension experiments. This cord was run over a pulley fixed to a support directly above, and to the distal end of the cord were fastened weights equal to those hanging on the plant. By this method, a weight above pressing down and another below pulling up, there was a compressed region; while the upper and lower parts of the stem were free from compression.

Variation in the method of hanging the weights on the plants was employed, but these details are of minor importance, and will not be given here.

*Helianthus annuus*

Twenty-six sunflowers were subjected to compression by the different methods explained above, the method depending upon the size and age of the plant used. Seedlings were cultivated in 20<sup>cm</sup> pots in the greenhouse. The duration of the experiments varied from two to five weeks. The weights varied from 400 to 2100<sup>gm</sup>. As a general result of this experiment one may say that a compression strain will call forth slightly increased mechanical development in a majority of the plants used. This expresses itself in a slightly greater thickness of cell wall and xylem cylinder. The phloem bundles are also slightly larger.

*Fuchsia speciosa*

Five small Fuchsia stems were subjected to a compression strain of 185<sup>gm</sup>. No bending was present to modify the anatomical structures. Sections were taken from the stems within the region of compression at distances from the ground varying from 5 to 8.5<sup>cm</sup>. Sections of the normal stems taken at corresponding places were selected for comparison. With the aid of an eye-piece micrometer, measurements of the xylem cylinders were made in two directions at right angles to each other. The average of these two measurements was taken as the average width of the xylem ring. Data so obtained indicated a greater thickness of xylem in the tension plant than in the control, but the increased mechanical development was not strong.

*Vinca major*

Eight young upright Vinca stems were subjected to compression by hanging the weights upon them. To prevent bending, the plants were tied to a support in the usual way, the final weight carried by each plant being 200<sup>gm</sup>. The period of experimentation extended from March 10 to March 25. Control plants were selected with reference to the same age, growth, and vigor. Sections taken in the region of stress were compared with sections taken at corresponding places in normal plants.

Observations under the microscope indicated, in a majority of the plants experimented upon, that a compression strain causes slightly increased development of mechanical tissue.

*Coleus tricolor*

Four young and thrifty Coleus plants were put under compression in a similar manner as explained for Fuchsia. After a period of twelve days under a compression strain of  $225^{\text{gm}}$ , they were gathered and together with their controls were placed in 50 per cent. alcohol for examination later.

Observations under a magnification of 150 diameters revealed no evidence of any differences between control and tension plants. We are hardly justified, then, in concluding that in these Coleus plants compression exerts any influence toward a development of mechanical tissue. For reasons that are apparent, the absolute amounts of xylem in both normal and tension plants were not determined by either the area or weight methods. The conclusion drawn was the result of detailed observations under the microscope by two persons working independently.

*Conclusion*

The foregoing experiments were performed to determine the influence of longitudinal compression upon the formation of mechanical tissue in the stems of *Helianthus annuus*, Vinca, Fuchsia, and Coleus. The results indicate that the stems of all the above-named plants except Coleus reacted to a compression strain by a self-regulatory development of mechanical tissue, yet the evidence cannot be called conclusive.

## C. INFLUENCE OF TENSION ON THE ROOT SYSTEM

A number of papers on the influence of pull, or tension, on stems, petioles, and other aerial organs have been published, but nothing as yet has reached print concerning the influence of this treatment on the root system. It is the purpose of this section of the paper to give the results of experiments along this line, and to show that in the root system we have an organ that reacts to tension by a self-regulatory development of mechanical tissue.

Tension on the root system was applied by means of a pull upon the stem, brought about in the usual way by weights, when the seedlings had grown to a height of about  $15^{\text{cm}}$  and the first pair of leaves had fully developed.

The seeds for any one series of experiments on *Helianthus annuus*

were chosen from the same head. No further selection was made. The *Ricinus* seeds were all taken from one variety. All the seedlings used for these experiments, except where otherwise indicated, were cultivated in an open field during the summer of 1905. The ground was carefully prepared with a mixture of clay, swamp muck, and manure. This was thoroughly spaded, and afforded a rich, firm soil, from which the seedlings were not easily uprooted when under tension.

Two series of experiments were run through, one in the earlier and the other in the later part of the summer. The duration of the experiments was generally two weeks, after which time the plants were dug up, together with their controls, and their root systems carefully washed and placed in 50 per cent. alcohol. The root systems of control and tension plants were first compared with reference to their external form and size and later with reference to their internal structures. The control plants were chosen from the same locality in which the plants under experimentation were grown and were selected with reference to similarity in size and vigor.

#### I. EXTERNAL ROOT FORM

To determine the influence of tension on the root system, a careful and close comparison was made between the control and tension plants. Special attention was given to observations upon the main and secondary roots. At times it was noticed that the lateral roots attained a greater development on account of the small or aborted growth of the main root. On the other hand, the main root would sometimes reach quite an enormous growth, in consequence of which the lateral roots would not attain the usual size. This great growth of the main root occurred far less frequently than that of the lateral roots. Thus what may be called a compensatory regulation holds true with reference to these two orders of roots. This compensatory regulation, then, must not be confused with the evident strengthening and increased development of roots due to tension. In our comparisons of the normal and tension plants this fact had always to be kept in mind.

The number, average length, and generally vigorous condition of the roots and rootlets were determined. It was found that in general the main root of the tension plant was straighter, tapered more

gradually, and attained greater length; the lateral roots were generally more numerous, larger in cross-section, straighter, and longer. The greatest growth of lateral roots was always found at the crown of the main root near the surface of the ground.

*Helianthus annuus*

To show the differences in root form between the normal and tension plants, two series of experiments were made on *Helianthus annuus*. The results of both were in general the same. All plants compared were of the same size and were grown in the open field under similar conditions. The first series was under experiment from July 8 to July 21, the second from September 21 to October 10. The final weights to which the plants were subjected varied from 750 to 1000<sup>gm</sup>.

The following are observations obtained from a comparison of the root systems of two seedlings of the same relative size and vigor. These were representative seedlings and the results obtained were characteristic of the entire set of thirty-four plants: The main root of the tension plant was straight for a distance of 6<sup>cm</sup>, that of the control plant undulate throughout its whole course. The main roots had the same diameter at the upper ends. At 5<sup>cm</sup> below the first lateral root, the diameter of the tension plant was 3<sup>mm</sup>; while that of the normal plant was 2<sup>mm</sup>. The normal plant had a cluster of 10 lateral roots at the upper end with a diameter of 1<sup>mm</sup> or more; while the tension plant had at its upper end a cluster of 13 lateral roots with a diameter of 1<sup>mm</sup> or more. All these were longer, much more branched, and in general appearance stronger than the lateral roots in the normal plant.

Here the differences between normal and tension plants were very marked, and it will be noticed later that marked differences appear as a result of microscopic observations.

*Ricinus communis*

The same general differences in external root form are noticeable in *Ricinus communis*. With this, as with the sunflower, two series of experiments were run through. The first was under experimentation from July 5 to July 21, and the second from September 26 to

August 11. The final weights held by the plants under tension varied from 1800 to 2000 gm.

The results of a comparison of two plants of *Ricinus communis*, selected with reference to similarity in all respects except in the matter of tension, are added below. The results here obtained are in general the same in the other sixty-seven plants used in the experiment.

The main root of the tension plant was straight throughout its whole course, that of the control undulate. The main roots had the same diameter at the upper ends. At 5 mm below the first lateral root the diameter of the tension plant was 4 mm, while that of the control was 3 mm. The control plant had a cluster of 18 lateral roots at the upper end with a diameter of 1 mm or less; while the tension plant had at its upper end a cluster of 23 lateral roots with a diameter of 1 mm or more. The lateral roots in the tension plant were much longer than those of the control plant. The root system of the tension plant was noticeably stronger in general appearance.

The results of these experiments on seedlings of *Ricinus communis* show how different the root systems of the plants under tension are from those growing under normal conditions. In every respect the differences are quite marked. Similar experiments conducted in the greenhouse with both *Helianthus* and *Ricinus* plants show like results.

From the above data one may conclude that tension causes a thickening and strengthening of the roots of *Helianthus annuus* and *Ricinus communis* grown in the open or in the greenhouse.

## 2. INTERNAL ROOT STRUCTURE

For a study of the internal root structure, freehand sections of the main and lateral roots were made. Sections of the main root were generally taken in two places, one near the top and the other some distance below. Sections of the lateral roots were made 1 to 2 mm from their insertion on the main root. The lateral roots to be sectioned were taken from the upper cluster of rootlets or from a position on the main root not lower than 1 cm from the first large lateral root. More than one lateral root on each plant was sectioned so as to obtain average results. The strongest and the average rootlets were always selected for comparison. No rootlet was chosen

unless it looked healthy, reached a fair length, and had a number of branches. Often the largest lateral root of the normal plant was not so large as the largest in the tension plant. This was sometimes taken as evidence of greater growth due to tension, but the results were not based on this fact alone. From the tension plant was selected a rootlet which was as large and vigorous as the largest rootlet in the normal plant. A comparison could then be made and the results, whatever they were, could be accepted without further allowance or additional estimation.

*Helianthus annuus*

Sections were taken of the main root at various points below the first lateral root. These were compared with sections taken at corresponding places in the control plant. The diameters of the sections of the normal root averaged 1<sup>m</sup>m greater, and for this reason a better mechanical growth would be expected in the control. As the result of careful observations the xylem was found to be greater in amount and the cells were thicker-walled in the tension plant than in the normal. In cases where the diameter of the sections of the normal and tension roots were the same, the tension root always showed the greater amount of xylem and thicker-walled cells.

From a comparison of the rootlets at the crown of the main roots of the tension plants with those from the corresponding place in the control plants, it was seen that the former had a larger size, and microscopical study emphasized this fact and showed that the xylem cylinder was larger and the walls of the cells thicker in the tension plants. Rootlets of average size taken from tension plants were compared with others of the same size, vigor, and growth taken from the normal plants. The comparison showed that the tension plants had a larger amount of xylem and cells with thicker walls.

To recapitulate, we may say that, out of five *Helianthus* plants under tension, the main roots of all show a better development of mechanical tissue than the main roots of five control plants of similar development. With reference to the lateral roots, the majority in the experimental plants showed a better mechanical development than similar rootlets in the control plants.

*Ricinus communis*

It may be said by way of summary that twenty-five out of the thirty-four *Ricinus* plants under tension in this series show a greater development of mechanical tissue in both the main and lateral roots than in the control plants. Sections of the primary roots show considerable differences, while the secondary roots show some but not so great differences in favor of the tension plants.

*Helianthus annuus*

The plants for comparison were chosen from the series of experiments in the open field during the latter part of the summer. The evidence obtained from observations upon the main roots of twelve seedlings of *Helianthus annuus* shows that nine reacted to tension by an increased development of mechanical tissue, while all the lateral roots examined showed a greater development of mechanical tissue in the tension plants.

## IV. Summary

In the foregoing work, of the five stems tested for the increase of mechanical tissue under the influence of longitudinal pull, only that of *Vinca* showed a response, and in this the increase was not great.

Pull in the direction of the longitudinal axis of the plant called forth a small increase of mechanical tissue in the main and lateral roots of *Helianthus annuus* and *Ricinus communis*.

Compression tension brought small increases of mechanical tissue in the stems of *Fuchsia*, *Vinca*, and *Helianthus*. By the same method Coleus gave no response.

The investigation, the results of which are here recorded, was carried out under the direction of Professor F. C. NEWCOMBE, to whom I wish to express my grateful thanks for kindly encouragement and helpful suggestions.

UNIVERSITY OF MICHIGAN,  
ANN ARBOR, MICH.

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PERIODICITY IN THE PRODUCTION OF THE SEXUAL  
CELLS OF *DICTYOTA DICHOTOMA*<sup>1</sup>

CONTRIBUTIONS FROM THE BOTANICAL LABORATORY OF THE  
JOHNS HOPKINS UNIVERSITY, No. 6

W. D. HOYT.

(WITH TWO CHARTS)

The antheridia of *Dictyota dichotoma* (Huds.) Lamour. were described in 1855 (1), but the motility of the spermatozoids was not discovered until 1896 (2).

In 1898 WILLIAMS (3) stated that the sexual cells of this alga are produced in fortnightly crops bearing a definite relation to the tides, and suggested that illumination is the factor determining the time of fruiting. In an excellent paper on the cytology of the gametophytic generation (4) the same author added a few details to his previous statements, and in 1905 (5) he gave a full account of the periodicity of *Dictyota* on the north coast of Wales, near Bangor, and discussed the factors which may determine this behavior. He also recorded a similar periodicity in this alga at Plymouth, England. At both places the time of production of the sexual crops bears a definite relation to the spring tides. The spring tides are, of course, those of greatest range, having the highest floods and the lowest ebbs, and occur every two weeks, about the times of the new and full moons. The neap tides are those of least range, having the lowest floods and the highest ebbs, and occur between the springs. A summary of records taken at Bangor for seven years shows that at this place the sexual crops of *Dictyota dichotoma* are generally initiated several tides before the least neap, and liberated from three to five tides after the greatest spring. They thus extend through parts of two sets of spring tides.

In his discussion of the factors which may regulate the periodicity, WILLIAMS assumes that the determining factor is one that varies with the alternation of spring and neap tides, and is most marked at

<sup>1</sup> An abstract of this paper was presented before Section G of the A. A. A. S., New York City, December 31, 1906.

the greatest spring tides. The effect of aeration, temperature, pressure, and light is considered. The first three are deemed negligible because plants in different situations subjected to different conditions in these respects show the same behavior in fruiting. Light, then, is the remaining condition which, with the alternation of spring and neap tides, varies most for the plants of all situations, and this is believed to be the determining factor for the following reasons. (1) At Bangor the development of each crop is rapid at first, when there is comparatively little water over the plants at low tide and much light is obtained. The development is retarded with the approach of neap tides when the light is weak, and is accelerated by the following spring tides when the available light again becomes strong (*chart 1*). (2) Whether liberation shall occur on the third or fifth tide after the greatest spring tide depends on whether the preceding spring tides have been great or small, thus giving more favorable or less favorable light conditions for the rapid development of the early stages. (3) The length of time taken by any crop to pass from initiation to liberation is directly proportional to the number of days intervening between one set of spring tides and the next; in other words, to the number of days when the available light is weak. (4) During October and November the light becomes less intense at the low water of spring tides, and both initiation and liberation are delayed several tides. (5) At Plymouth, England, the crops seem to be produced wholly within one set of spring tides, and the time of liberation seems to be determined by the height of the tides during development. The light is less advantageous at the low water of spring tides, and liberation does not occur until seven to twelve tides after the greatest spring.

These facts seem to show a causal relation between the amount of available light and the time of fruiting, but a single experiment tends to make this conclusion less certain. Two sexual plants of *Dictyota* were brought into the laboratory in October and left undisturbed during the winter. When examined the following April, they produced three crops at fortnightly periods, although they had been removed from the alternating influence of the tides for six months. WILLIAMS therefore concludes that "periodicity of the sexual cells is an hereditary character and consequently may be expected to manifest itself in

seas and habitats where there are no tides. At the same time, the sexual cells are so responsive to changes in the amount of illumination that the time of their development in seas where there are tides is regulated by the increased illumination obtained during the low water of spring tides" (5, p. 548).

During a stay at the laboratory of the Bureau of Fisheries at Beaufort, N. C., the past summer, a record was kept of the fruiting of the sexual plants of *Dictyota dichotoma*, together with observations on the tides and the conditions of growth. The following facts were observed. (1) The sexual cells were produced in regular periodic crops. (2) The time of the production of these crops bore a definite relation to the tides. (3) The crops were borne at monthly instead of fortnightly intervals, appearing only at alternate spring tides. (4) Rudiments of sexual organs were first observed on the day of, or the day before, the greatest spring tide, instead of a few days before the least neap as at Bangor. (5) The time taken for the development of an entire crop was about eight or nine days, instead of twelve to seventeen days. (6) General liberation occurred six days after the greatest spring tide, instead of two to five days. (7) Differences in the height of different sets of spring tides had no effect on the time of liberation or the time taken for the development of an entire crop. Thus, the August crop was liberated six days after the greatest spring tide, the preceding ebb tides having been the lowest observed during the summer. The October crop was liberated at the same interval, although the tides for nearly a month previous had been exceptionally high at both ebb and flood. (8) Differences in the time intervening between one set of spring tides and the next had no effect on the time of initiation of rudiments or of liberation of gametes. Although the number of days between one spring tide and the next varied from twelve to eighteen, rudiments were always first observed about the day of the greatest tide, and liberation always occurred six days after the greatest tide of each alternate set of springs.

A comparison of the record of a single crop at Beaufort, beginning with the day when rudiments of sexual organs were first observed, with a similar record for a crop on the coast of Wales, will best illustrate the difference in development (*charts 1 and 2*). This record is followed in each case with male plants, and the numbers given are

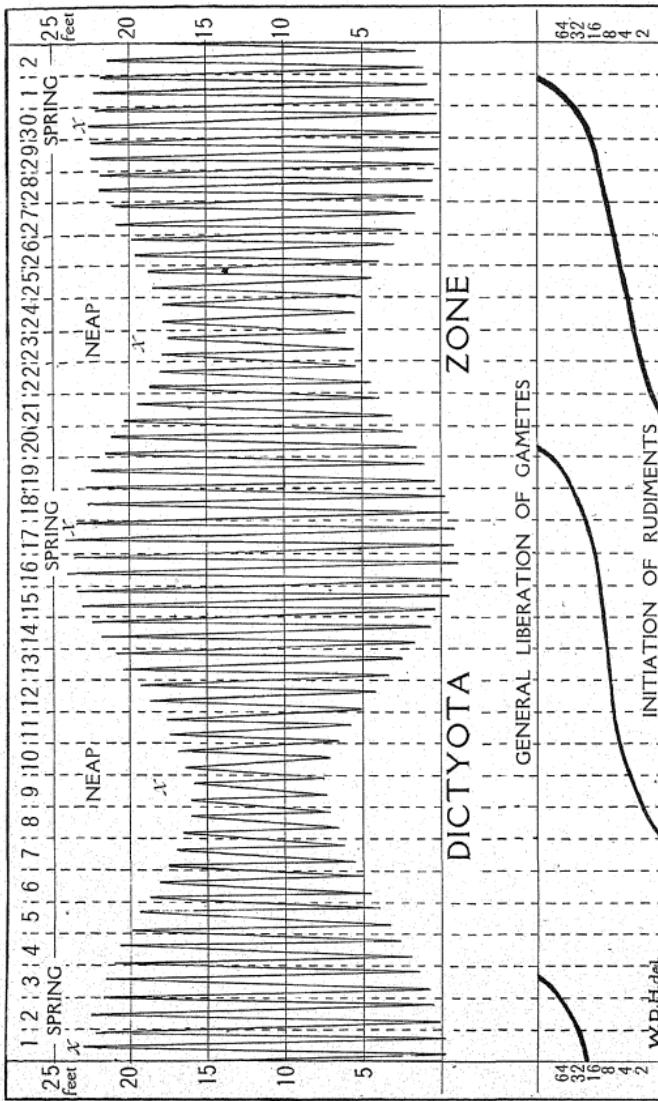


CHART 1.—Tidal relations at Bangor, Wales, for thirty-two days, showing great tides and two crops of *Dictyota*.

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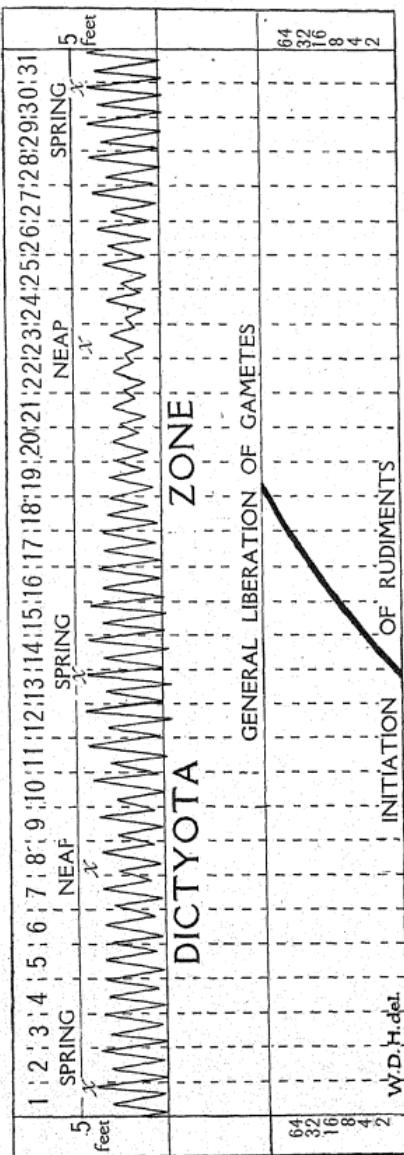


CHART 2.—Tidal relations at Beaufort, N. C., for thirty-one days, showing slight tides and only one crop of *Dictyota*.

The tide records are taken from the Tide Tables for 1906 of the U. S. Coast and Geodetic Survey. While wind and other factors may cause any tide to vary from the heights given at either Bangor or Beaufort, the figures show the general course of the rise and fall at the two places. The curves showing the development of *Dictyota* at Bangor are adapted from WILLIAMS (5, p. 559). The two charts are drawn to the same scale. The zig-zag lines show the rise and fall of the tide and the heights of the water in relation to the *Dictyota* zone. The curved lines show the development of the sexual crops of *Dictyota* for the respective periods with their relations to the tides. The numbers opposite the crop curves indicate the number of cells seen in the surface-view of each antheridium.

those of cells seen in the surface-view of each antheridium. The actual number of cells in each antheridium is much greater. These show as follows:

	BEAUFORT	BANGOR
1st day.....	Undivided rudiments	Undivided rudiments
3d day.....	8-16 cells	" "
4th day.....	32-64 cells	" "
5th day.....	64 cells	" "
6th day.....	General liberation	
7th day.....	Liberation from belated sori	2-4 cells
10th day.....		Many 64 cells
12th day.....		General liberation
13th day.....		Liberation from belated sori.

In addition to this difference is the fact that the records taken make it probable that every crop produced during the fruiting season at Beaufort was similar in development to the one described, while at Bangor there is considerable difference in the development of different crops. The cause of these differences in the fruiting of the plants on our coast and on the coasts of Wales and England is not apparent. A comparison of the tides at the different localities, however, is of interest in this connection (6) (*charts 1 and 2*).

The average range of the tide is: near Bangor, Wales, 17.9 feet; at Plymouth, England, 11.5 feet; at Beaufort, N. C., 2.8 feet. The average difference in the height of low water at spring and neap tides is: near Bangor, Wales, about 5.7 feet; at Plymouth, England, about 3.4 feet; at Beaufort, N. C., 0.5 foot. We are still unable to say whether these differences will explain the difference in behavior, or whether this is due to some other, possibly ancestral, cause.

It is possible that the *Dictyota* occurring at Beaufort is a different species from that found at Bangor or Plymouth. To determine this, a specimen was sent to Mr. FRANK S. COLLINS of Malden, Massachusetts, who verified the identification, referring it to *D. dichotoma*. In spite of the morphological resemblance, however, it is possible that the Beaufort form is a different physiological species, and this can be determined only by means of cultures. The final test can be made only by growing some of the Bangor plants at Beaufort and *vice versa*. It is hoped that this can be done at some future time.

The explanation of this periodicity is not apparent. Although the

behavior of this alga at Bangor and Plymouth may be explained by the assumption that illumination is the determining factor, the facts observed at Beaufort indicate that for the plants of this region, at least, some other explanation must be sought. That the time of fruiting is determined to some extent by the tides is indicated by the very exact relation which this bears to the tides. That illumination is not the determining factor is indicated by the facts that fruiting occurred only on alternate spring tides, that differences in the amount of light as determined by the state of the weather and the heights of different sets of spring tides had no effect on the time of fruiting, and that the differences in light conditions to which individual plants are subjected are greater than those differences to which all the plants are subjected because of differences in the height of water at spring and neap tides.

As was mentioned above, the average difference in the height of low water at Beaufort at spring and neap tides is only 0.5 foot. As the difference in the depths at which individual plants grow is at least six times as great as this, the difference in the amounts of light received by different plants at any instant is greater than the differences in the amounts of light received by any one plant at low water of spring tide and at low water of neap tide. And yet all the sexual plants observed in all situations fruited at the same time. It may be urged that the sexual plants are sensitive, not to the actual amount of light, but to the maximum intensity, and that this occurs at the time of the spring tides for the plants of all situations. But the same reasoning applies to all the other factors which are influenced by the tides—such as aeration, pressure, etc.—so that there is no more reason on this ground to ascribe the behavior to the light than to any one of the other factors.

One observation is interesting in this connection. A female plant was brought into the laboratory August 11 and placed in a covered jar of sea-water near a window. As is often the case when *Dictyota dichotoma* is subjected to unfavorable conditions, proliferations were formed from the margin of the thallus. When examined October 6, most of the plant had died, but several proliferations were still alive, and these showed sori of uniform age only slightly less advanced than those on plants growing in the harbor. In this case

the entire plant had been removed from alternating conditions (except those of day and night) for nearly two months, and the sori were borne on proliferations produced while the plant was in the laboratory, consequently on parts of the thallus which had never been subjected to alternating conditions. This result is similar to one obtained by WILLIAMS (5, p. 547), but goes one step farther in showing that the periodicity is transmitted to new structures formed from the original plant, even when these are not subjected to alternating conditions. These proliferations were, of course, produced by the protoplasm of the parent plant; it still remains to be seen whether the periodic habit can be transmitted through the protoplasm of the egg.

A few observations were made at Beaufort in the summer of 1905, but no satisfactory records were obtained. The observations made, however, indicate that the behavior of the sexual plants was like that observed during the past summer.

Many specimens of several species of *Dictyota* have been obtained from two localities in Jamaica. Among these are four sexual plants—two male and two female—collected by the author at Port Antonio, April 13, 1906, and eight sexual plants—two male and six female—collected by Mr. I. F. LEWIS at Port Morant, July 16, 1906. Although this material is too scanty to warrant a conclusion, it is interesting to note the direction in which the evidence points.

The specimens from Port Antonio have all the sori of about the same age, and at the same stage of development that would be expected for plants of *Dictyota* at Beaufort, N. C., on the same day. In the specimens from Port Morant, the male plants have all the sori of the same age, while all the female plants have sori of two ages, one set being mature, and the other being young and at a stage corresponding to that of the antheridial sori on the male plants. These sori of two ages probably represent two crops of eggs, the older of which was not shed before the younger one was produced. The age of the older crop cannot be stated with certainty, but the age of the younger crop of eggs and of the antheridial sori does not agree with the age of sori on plants collected by the author on the same day at Beaufort. They seem at about the stage of development, however, that would be expected for *Dictyota* at Bangor on this day. These facts indicate that, on the coast of Jamaica also, *Dictyota* shows periodicity in the production of sexual cells. As there are practically no tides on this

coast (about six inches), this condition, if true, supports WILLIAMS' conclusion that "the periodicity of the sexual cells is an hereditary character and consequently may be expected to manifest itself in seas and habitats where there are no tides."

As the Jamaican specimens of *Dictyota* belong to more than one species, the facts indicate the probability that periodicity obtains throughout the genus. The apparent correspondence in the time of fruiting of the Port Antonio specimens with those of Beaufort, and of the Port Morant specimens with those of Bangor may be only a coincidence. It is certain, however, that the time of fruiting of the Port Morant specimens did not coincide with that of *Dictyota* at Beaufort. Evidently we need more observations before we can draw general conclusions or suggest explanations of the facts already observed.

The author is able to verify WILLIAMS' statements that light is not necessary for the escape of the sexual cells of *Dictyota dichotoma*, and that the tetrasporic plants show no periodicity.

From a single set of observations, WILLIAMS gives evidence of periodicity in "Haliseris" and suggests by inference the possibility of its occurrence throughout the Dictyotaceae. A species of *Padina*, probably *P. Durvillaei* Bory, is abundant at Beaufort in the same situations with *Dictyota*. Numerous male plants were found and these showed no evidence of periodicity, sori of all ages occurring on the same plant. It seems, therefore, that periodicity is not characteristic of all the Dictyotaceae. All the sexual plants of *Padina* which were examined bore only antheridia and produced these on both sides of the thallus, differing in these respects from the printed descriptions. The antheridia were numerous, being borne in sori in wide concentric rows which appeared white to the naked eye, and the sori were usually produced in regular acropetal succession.

The author expects to continue this work, but, believing the present data to be insufficient to explain this periodicity, gives the results already obtained with the hope of inducing other botanists who are favorably situated to begin a study of this form. It seems that only by observations of the fruiting and careful comparison of the conditions at a number of places for a considerable period of time can we hope to understand this striking phenomenon.

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the Fisheries Laboratory at Beaufort, N. C.; to Mr. H. D. ALLER, Acting Director of the Fisheries Laboratory at Beaufort, N. C., for many courtesies extended during this investigation; to Mr. FRANK S. COLLINS, Malden, Massachusetts, for verifying the determination of the species of *Dictyota* occurring at Beaufort; to Dr. MARSHALL A. HOWE, New York Botanical Garden, for information concerning *Padina*; to Mr. I. F. LEWIS, Johns Hopkins University, for specimens of *Dictyota* from Port Morant, Jamaica; and to Professor D. S. JOHNSON, Johns Hopkins University, for helpful criticisms and suggestions.

#### CONCLUSIONS

1. As on the coast of Wales and England, *Dictyota dichotoma* at Beaufort, N. C., produced its sexual cells at regular intervals, bearing a definite relation to the tides.
2. The time of production of these crops, however, differs from that on the coasts of Wales and England, the crops being borne at monthly instead of fortnightly intervals.
3. The assumption that light is the sole factor determining the time of fruiting does not hold for the plants of *Dictyota dichotoma* growing at Beaufort.
4. Specimens of *Dictyota* from Jamaica indicate that there also the sexual cells are produced in periodic crops, and that periodicity may obtain throughout the genus. The time of fruiting, however, at least in some cases, is different from that of *Dictyota* at Beaufort.
5. Periodicity in the production of the sexual cells is not universal among the Dictyotaceae. A species of *Padina*, probably *P. Durvillei* Bory, bears antheridial sori of all ages on the same plant.

JOHNS HOPKINS UNIVERSITY  
BALTIMORE

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## THE FUNCTION OF INVERTASE IN THE FORMATION OF CANE AND INVERT SUGAR DATES

A. E. VINSON

The chemical character of the ovulary of the date, *Phoenix dactylifera*, has seldom been studied, although the seed has long furnished a convenient and favorite material for the most diverse researches. Published analyses with few exceptions have been made by the usual WEENDE methods and consequently give no idea of the real chemical nature of the fruit. A French analysis<sup>1</sup> made before 1867 mentions

	Per cent.
Water	43.6
Albuminoid and pectin bodies	2.9
Gallic acid and glucose	47.9
Inulin	trace
Fat	0.4
Cellulose	1.9
Mineral matter	3.3

a trace of inulin, but otherwise is of little significance.

SENY<sup>2</sup> seemed to strengthen the prevalent view, that dates contain little or no cane sugar, by the analysis of a number of Mesopotamian dates which contained dextrose (invert sugar) only. In reply to SENY's paper, however, LINDET<sup>3</sup> points out, that a sample of Tunis dates—probably Deglet Noors which occur on the Paris market—analyzed by him in 1891 contains 23 per cent. of glucose (invert sugar) and 38 per cent. of cane sugar. This is the first mention of a cane sugar date that I have found, aside from recent comparative analyses, made at this station, of Arizona-grown dates from trees imported from all the date-growing regions of the world by the U. S. Department of Agriculture and placed in the Cooperative Date Orchard at Tempe, Arizona.

LINDET cautions against generalizing from a few analyses when they concern plants and fruits, for the absolute and relative proportions of sugars will vary with climate, condition of season, degree of maturity, etc. In this paper I intend to show that other factors,

<sup>1</sup> Recueil de Mémoires de Médecine de Chirurgie et de Pharmacie Militaires.

<sup>2</sup> Journal de la distillerie française. 1895.

<sup>3</sup> Ibid. 118. 1895 [abstract Chem. Zeit. 20:898].

especially the presence of invertase, and probably limitations in the distribution of the same, are more potent in determining the relative proportion of the sugars than the factors mentioned by LINDET. According to observations on the date, it seems that fruits of the same species but of different chemical nature are true chemical mutants, determined by the plant itself rather than by its environment. Further investigations may reveal cane sugar varieties of what BUGNET<sup>4</sup> considered inverts sugar fruits, as grapes, currants, and figs, or invert varieties, among the partially cane sugar fruits, bananas, apricots, peaches, plums, apples, and pears.

SLADE,<sup>5</sup> basing a classification on the analyses of varieties, from the Tempe orchard, of various native seedlings and of native fruits from Mexico, divided dates into two main classes: cane sugar dates, embracing the Deglet Noor, and M'Kentichi Degla; and invert sugar dates, embracing Rhars, native seedlings, Mexican dates, and most others. It is possible to have partial cane sugar dates, and such do exist, as I shall point out later. While SLADE's classification is correct for mature and cured dates, nevertheless all dates are decidedly of the cane sugar type at some time of their life-history.<sup>6</sup> This fact gave the first clue to the influence of invertase in determining the saccharine character of the date and probably of most other fruits.

It appears probable that the carbohydrates may enter the fruit as cane sugar, for nearly all fruits, either cane or invert sugar, show an appreciable percentage of cane sugar at the period of maximum accumulation of dry matter. This is often very inconsiderable, as we see in KEIM'S<sup>7</sup> analysis of ripening cherries, and may not be detected at all, as in the ripening persimmon.<sup>8</sup> Even in these cases more cane sugar would be found if special precautions were taken to destroy the invertase immediately after removing from the tree. The cane sugar of the date vanishes very rapidly in the invert varieties. MIERAN<sup>9</sup>

<sup>4</sup> Compt. Rend. 51:894. 1860; through Bull. 94, Bur. of Chem., U. S. Dept. Agric.

<sup>5</sup> All notes on the unfinished work with dates of H. B. SLADE (died June 5, 1905) were turned over to the writer and acknowledgment is made in this paper whenever material from that source has been used.

<sup>6</sup> Ann. Rep. Arizona Agr. Exp. Sta. 17:164, 165.

<sup>7</sup> Zeit. Anal. Chem. 30:401. 1891.

<sup>8</sup> BIGELOW, CORE, and HOWARD. Jour. Am. Chem. Soc. 28:688.

<sup>9</sup> Chem. Zeit. 56:1003, 1021, 1283. 1893.

found also that banana pulp not only reduced its own cane sugar rapidly, but could invert large amounts of added cane sugar. BAILEY,<sup>10</sup> however, states that he failed to find any enzymic action in the ripening of the banana, but his figures show a dropping-off of cane sugar after the yellow-brown stage.

In a set of tests for invertase with very green, yellow, and black-ripe bananas, made in this laboratory, a feeble inverting power at room temperature was found in the two latter conditions, but not at all comparable with that of an invert sugar date under like conditions. It is possible that cane and invert sugar bananas may be found. In the banana, however, the reserve material in the fruit itself is largely starch and is finally converted into cane sugar, either directly or by way of maltose. Nevertheless, it would seem that some cane sugar, as such, may come into the banana ripened on the tree, for both BUIGNET<sup>11</sup> and RICCIARDI<sup>12</sup> agree that when ripened naturally this fruit contains more cane sugar and less invert sugar than when ripened artificially. Substantially the same is true with the cane sugar date. It is not untenable, however, that some other soluble carbohydrate may enter the fruit and be rapidly synthesized to cane sugar, for BROWN and MORRIS<sup>13</sup> have shown that the excised barley embryo, cultivated on maltose solution, formed cane sugar very rapidly by the "action of the living embryo cell." They found also that living embryos did not synthesize cane sugar from glucose. This difficulty is not insurmountable, for HILL<sup>14</sup> demonstrated the reversible nature of enzymic action by synthesizing maltose from glucose in very dilute solution by the aid of maltase. The maltose formed was later found to be isomaltose,<sup>15</sup> but that does not necessarily mean that, under cell conditions, true maltose would not be formed. The reverse action of enzymes has been shown to be influenced, at least with regard to speed, by the presence of a third body. Thus ethylbutyrate is formed more rapidly by lipase in the presence of lecithin.<sup>16</sup>

<sup>10</sup> Jour. Biol. Chem. 1:355. 1906.

<sup>11</sup> Compt. Rend. 49:276-278. 1859; through Bull. 94, Bur. of Chem.

<sup>12</sup> Compt. Rend. 95:393. 1882; *ibid.*

<sup>13</sup> Jour. Lond. Chem. Soc. 57:458 (517). 1890.

<sup>14</sup> Jour. Lond. Chem. Soc. 73:634. 1898.

<sup>15</sup> Through LOEB, Dynamics of living matter 11. New York 1906.

<sup>16</sup> HEWLETT, *ibid.* p. 10.

That cane sugar may find its way directly into the fruit would seem probable, moreover, since it is one of the most diffusible carbohydrates, and BROWN and MORRIS<sup>17</sup> believe it the primary carbohydrate synthesized by the chloroplasts. Cane sugar, however, is not suited to nourish the protoplasm. If injected directly into the blood of animals, BERNARD<sup>18</sup> has shown it to be eliminated unchanged by the kidneys, and to be inverted in the beet root before being used. Glucose appears in the beet root and can be traced up the stem, but not so with cane sugar. FORBES<sup>19</sup> finds the same true with canaire, *Rumex hymenosepalus*. The small amount of cane sugar contained in the dormant root suddenly decreases when the root begins to send up a stalk. The much larger amount of starch is similarly affected. As is well known, the moving carbohydrate of the maple is nearly pure cane sugar. Plant leaves are also known to contain invertase in conjunction with cane sugar. BROWN and MORRIS consider the entire amount of cane sugar to be hydrolyzed, first for the nutrition of the tissues, and then any excess to be resynthesized into starch for future use, the hydrolytic product of this starch being maltose. It appears thus that either cane sugar, invert sugar, or maltose has the opportunity of leaving the leaf. SAPOSCHNIKOFF<sup>20</sup> says the form in which the carbohydrate leaves the leaf is unknown, but is probably glucose.

It has been established, on the other hand, that the existence of starch in the tuber of the potato,<sup>21</sup> in maize,<sup>22</sup> and in wheat<sup>23</sup> depends on the previous existence of cane sugar in the juice of these plants. KEMI,<sup>24</sup> in HILGER's laboratory, found cane sugar to accumulate in the leaves of the cherry and starch in the fruit stems during the growing stage, both of which disappear at the period of maximum ripening, when a small amount of very transient cane sugar is found in the

<sup>17</sup> A contribution to the chemistry and physiology of foliage leaves. Jour. Lond. Chem. Soc. 63:604. 1893.

<sup>18</sup> GREEN, The soluble ferments 110. Cambridge 1899.

<sup>19</sup> Unpublished papers.

<sup>20</sup> Bildung und Wanderung der Kohlenhydrate in der Laubblättern. Ber. Deutsch. Bot. Gesells. 8:233. 1890.

<sup>21</sup> GIRARD, Compt. Rend. 108:602. 1889.

<sup>22</sup> ZEPLEY, Compt. Rend. 94:1033. 1882.

<sup>23</sup> BALLAND, Compt. Rend. 106:1610. 1888.

<sup>24</sup> Loc. cit.

fruit. They explained this erroneously by considering that the acidity of the leaf was insufficient to invert the cane sugar, and only after it reached the acid fruit could such inversion take place. The occurrence of some cane sugar in the earliest stages of growth of the cherry, and its subsequent disappearance, they also attribute to a lack of acid. BUIGNET<sup>25</sup> attempts to explain the high cane sugar content of some strawberries in much the same way, on the hypothesis that the sugar and acid exist in different cells, and that in the watery varieties diffusion and consequently inversion take place more rapidly than in drier berries. In a later paper<sup>26</sup> he concludes that acidity has nothing to do with the inversion, but that it is probably the work of a "nitrogenous ferment." In the date acidity has nothing to do with inversion, for the cane sugar dates are usually the more acid. Neither does the high acidity of the orange appear to determine inversion, for here the cane sugar increases during ripening, while the invert sugar remains nearly constant. This BERTHELOT and BUIGNET<sup>27</sup> think is especially remarkable, since the green fruit contains no starch from which to derive the cane sugar. The phenomenon, however, is almost identical with that of the cane sugar date and just what would be expected of any fruit having similar invertase relations. The accumulation of starch in the stem of fruits which contain none appears to be quite common and has been observed in the grape.<sup>28</sup> Whether or not this is the case with the date has not been determined.

While the observed fact that cane sugar accumulates at the time of maximum ripening in nearly all fruits, even the pea<sup>29</sup> and cucurbits,<sup>30</sup> points very strongly to that sugar as the original carbohydrate to enter the fruit, one point militates most powerfully against such a hypothesis; the partial osmotic pressure of cane sugar within the date would stop flow in that direction long before the high observed

<sup>25</sup> Résumé, Compt. Rend. 49:276-278. 1859; through Bull. 94, Bur. of Chem.

<sup>26</sup> Compt. Rend. 51:894. 1860; *ibid.*

<sup>27</sup> Compt. Rend. 51:1094. 1860; *ibid.*

<sup>28</sup> FAMINTZIN, Ann. Oenol. 2:242. 1871; and HILGER, Landw. Vers. Station 17:245. 1874; through KEIM, *loc. cit.*

<sup>29</sup> SCHWARZ AND RIECHEN, Zeits. Unters. Nahr. Genuss. 1904:550; and FREERICHS AND RODENBERG, Arch. Pharm. 243:276. 1905.

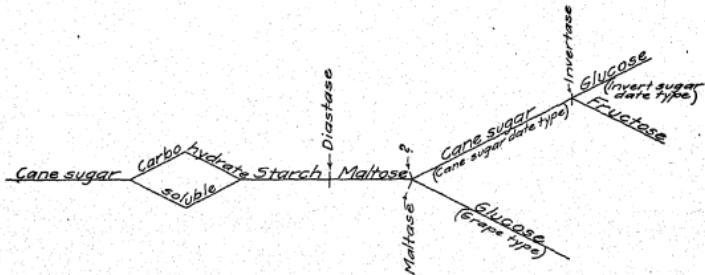
<sup>30</sup> LECLERC DU SABLON, Compt. Rend. 140:320, 321. 1905.

percentages of cane sugar could be approached. The same is true also for glucose and fructose, for on the inversion of the cane sugar present they appear in approximately molecular proportions as determined by BROWNE's<sup>31</sup> method. Occasional samples vary considerably from this relation, giving sometimes a large excess of fructose, sometimes of glucose. This may be explained in part by a number of possibilities. The difficulties of technic in taking consecutive representative samples of such material as pulped date must be taken into consideration because data from two different samples enter the BROWNE formula. An excess of fructose may be accounted for by the hydrolysis of inulin. SLADE found inulase in some dates but has left no record of having found inulin. Neither inulin nor inulase was found by the writer in a sample examined in that regard. An excess of fructose over glucose might also originate in the preferential use of glucose by the tissues, as is known often to be the case. An excess of glucose might come from maltose and SLADE observed maltase, but I have not yet examined the date for that enzyme. Here again an excess of glucose might be consumed by the tissues, leaving nearly pure invert sugar. The relation between fructose and glucose in other fruits has never been worked out, but it appears that in the case of the date nearly all the sugar is cane sugar or a derivative of cane sugar.

Times thus undoubtedly occur with every individual when the partial osmotic pressure of these three sugars is approximately equal; nevertheless, accumulation of total sugars goes on rapidly. The date, then, as a species at least, must be prepared to pass in carbohydrate against a relatively high osmotic pressure of cane sugar, glucose, or fructose, either singly or simultaneously. Unless we choose to hold that the intercellular threads of living protoplasm pass along colloids and crystalloids against osmotic pressure, we are forced to seek some likely soluble carbohydrate against which there exists no considerable pressure. This place seems to be filled by maltose. So far as I am aware, neither the presence nor the absence of maltose has been demonstrated in the growing date. We should never expect to find any great amount, and it might be so transient as to escape detection entirely.

<sup>31</sup> Bur. of Chem. Bull. 90:10; also Jour. Am. Chem. Soc. 28:439. 1906.

The following scheme for the translocation of sugars from stems and leaves to fruit seems then to be in accord with all known facts and contradicted by none. In the early stages cane sugar or mixed sugars move into the fruit until osmotic equilibrium is reached. After that, excess of cane sugar may be stored as starch within the fruit, as in the case of the banana and apple, thus diminishing the osmotic pressure for that sugar; or a part may be reduced to invert sugar, farther reducing the cane sugar pressure. On ripening, the starch is again transformed to soluble sugars. In those cases where no starch occurs in the fruit itself, but in the stem, the latter is probably a laboratory and not an ordinary storage tissue. The plant would scarcely have found it advantageous to thrust storage duties on the



stem when the fruit was fully capable and destined to receive the same material later. It seems more probable that after osmotic equilibrium is reached between the fruit and sap, those sugars against which there is osmotic pressure would be changed to starch at the threshold, and this starch in turn hydrolyzed by an amylase into maltose. Maltose, against which there is little or no osmotic pressure, would enter the fruit and there be split up into glucose or rearranged into cane sugar, which would be inverted where it comes into contact with invertase. Probably all these reactions take place to a greater or less degree in any ripening fruit, but the predominance of one or the other leads to a special type. If the main reaction were maltose to glucose, fruits of the grape type would result. If maltase secretion were suppressed, the maltose would follow the transformation it takes in fruits of the date type.

It is true that no catalytic outside the living cell is known which will transform maltose into cane sugar, but the cell is known to do so and the catalytic will probably be discovered. The transformation of starch to cane sugar seems to be established for the banana and apple, but here again no catalytic is known that will cause the transformation, although many, and some of organic origin, transform it into maltose. BROWN and MORRIS, moreover, have shown maltose to be the final product of starch hydrolysis in the leaf. Thus it seems probable that in these cases starch changes to cane sugar through maltose. It is also probable that the speed of transformation, maltose to sucrose, is very considerable, since BIGELOW and GORE<sup>32</sup> find the curve representing the increase of cane sugar in a ripening apple to be nearly the reverse of that representing the decrease in starch.

An examination of the accompanying table will reveal the chief points of interest in the development of the date, and also the divergence between the cane and invert sugar varieties. The cane sugar date is represented by the fruits of the Deglet Noor<sup>33</sup> from palms imported from the oasis of French Algeria in 1900. The invert sugar date is from a seedling palm of excellent quality growing upon the university campus and immediately accessible from the laboratory.

It seems that any attempt to interpret a series of date analyses, made at different stages of growth, by reducing them to the dry basis, can give only a partial idea of what is actually taking place. Water has as much to do with the changes as cane sugar or invert sugar. A date having 20 per cent. of dry matter may change in a few days to one of 60 per cent. in three ways: by the loss of water through transpiration with decrease in weight; by the addition of dry matter without loss of water, possibly some gain, and increase in weight; or by the addition of dry matter with loss of water and little or no change in weight. In the case of the cherry, KEIM has noted that the period of maximum increase in percentage of dry matter in the fruit is parallel with that of maximum increase in weight. In the peach<sup>34</sup> the increase in solids is fairly proportional to the increase in water. With the date there is a more or less parallel increase in percentage of dry matter and

<sup>32</sup> Bull. 94, Bur. of Chem., U. S. Dept. Agric.

<sup>33</sup> SWINGLE, Bur. Plant Ind., U. S. Dept. Agric. Bull. 53:33.

<sup>34</sup> BIGELOW and GORE, Bur. of Chem., U. S. Dept. Agric. Bull. 97:12.

## CANE SUGAR DATE; DEGLET NOOR—TEMPE, ARIZ.

Date	Condition of sample	Condition of bunch	Av. wt. grams	Av. wt. seeds	Per cent. dry matter	Per cent. cane sugar	■ Per cent. fructose in cane sugar
Sept. 18, 1906	Pea size.....	Belated.....	1.15	0.145	1.25	1.99	2.62
" 18 " 1906	Green, about half grown.....	" .....	5.02	1.572	17.41	2.82	3.34
" 18 " 1906	Slightly yellow.....	" .....	5.02	0.905	20.15	2.90	3.20
" 18 " 1906	Green, half grown.....	" .....	6.96	0.905	10.58	2.84	4.17
Oct. 5, 1906	Green, yellow tinged.....	" .....	6.66	0.705	17.22	3.78	3.76
" 5 " 1906	Red.....	" .....	8.32	0.668	11.77	2.06	3.76
" 5 " 1906	Red, turned green to red.....	" .....	9.08	1.079	10.82	3.54	4.39
Sept. 7, 1906	Light green, more marked.....	" .....	6.37	0.902	15.10	3.09	3.97
" 7 " 1906	Light green, red at tip.....	" .....	6.40	0.704	23.59	2.80	5.46
Oct. 17, 1905	Bright yellow.....	" .....	7.19	0.911	12.66	2.96	3.49
" 17 " 1905	Red, yellow at base.....	" .....	8.84	0.656	12.57	2.93	2.04
" 17 " 1905	Red, stored 10 days.....	" .....	8.45	0.650	12.66	3.36	3.49
" 17 " 1905	Red, stored 10 days.....	" .....	8.45	0.638	7.55	2.16	2.55
" 17 " 1905	Red, considerately yellow.....	" .....	8.45	0.638	7.55	2.16	2.55
" 17 " 1905	Slightly spotted.....	" .....	8.32	0.671	7.33	3.45	5.40
" 17 " 1905	About one-third soft.....	" .....	8.32	0.674	7.22	3.11	5.99
" 27 " 1905	Slightly dry, very soft.....	" .....	8.35	0.674	7.08	5.57	5.57
" 27 " 1905	Apparently fully ripe.....	" .....	8.35	0.612	59.60	6.56	8.86
" 31 " 1905	Art., ripened 30 hours from red.....	" .....	8.95	0.656	62.65	33.07	6.86
" 31 " 1905	Same apparently inferior.....	" .....	7.48	0.639	63.60	35.00	12.39
" 31 " 1905	Art., ripened 5 days from red.....	" .....	7.48	0.639	63.64	21.09	14.13
Oct. 10, 1906	One-half to two-thirds ripe after 5 days.....	" .....	7.48	0.637	63.62	21.18	14.12
Nov. 13, 1905	Ripe on tree.....	" .....	9.00	0.732	8.13	62.49	7.19
" 13 " 1906	Ripe on tree.....	" .....	7.32	0.512	7.83	60.58	10.03
" 13 " 1906	Art., ripened, stored 21 days.....	" .....	7.36	0.512	74.01	31.51	8.31
Dec. 4, 1906	Ripe on tree after frost.....	" .....	6.35	0.626	11.36	69.31	13.41
			8.49	0.643	8.44	73.32	44.71

## CANE SUGAR DATE; DEGLET NOOR—HEBER, CALIFORNIA

	Invert sugar date; seedling—campus, Tucson, Ariz.		
Nov. 14, 1906	Ripe on tree.....	0.50	0.800
Dec. 10, 1906	Finished in incubator.....	10.54	0.788
		10.54	0.788
Sept. 7, 1906	Very green.....	10.56	1.215
Oct. 3, 1906	Yellow, after cold week.....	10.67	1.100
" 10 " 1906	Yellow, ripe in spots.....	11.26	1.084
" 10 " 1906	Partly ripe, softened slightly.....	12.37	1.093
" 10 " 1906	Partly brown.....	12.18	1.093
" 10 " 1906	Ripe, not overripe.....	11.58	0.767
" 10 " 1906	Ripe, still light brown.....	12.07	0.907
Nov. 6, 1906	Ripe, stored 14 days.....	10.07	0.927
" 6 " 1906	Ripe, stored 1 week.....	8.21	0.709
" 6 " 1906	Ripe, stored 1 week.....	9.34	0.767

in weight (or size) till something like 20 per cent. of dry matter is present. Growth, so far as size is concerned, has then about reached its limit, but accumulation of dry matter in the form of sugar now takes place more rapidly, and in a relatively short time the dry matter rises from 20 or 25 per cent. to nearly 60 per cent. During this time there appears to be little or no further increase in weight. The rational interpretation then is that water is replaced by sugar. Curing again increases the dry matter up to 70 or even 85 per cent., but this change is purely a loss of water. These observations are further confirmed by purely practical ones. It may be observed that the individual dates on a bunch develop similarly throughout; the ones near the tips of the sprays remaining always somewhat smaller. At a certain period the bunch as a whole appears much the same and individuals begin ripening, but it may be a month or more before others on the same bunch, even on the same sprays, are ripe. The same thing is observed in artificial ripening experiments. Two sprays from the same bunch, and looking very much alike, will ripen under artificial conditions very differently. Some speedily develop into a plump, luscious fruit, while others dwindle away and finally furnish a thin skin of poor-quality flesh over an apparently normal seed. The seed seems to mature before marked changes in the ovulary begin. The fact that increase in dry matter takes place after the apparent maturity of the fruit must lie at the foundation of all economic attempts at artificial ripening. Any plans which ignore this will necessarily prove futile. The ripening process in the date, unlike that in the banana, is essentially one of addition and not of transformation. By reducing results to the dry basis, the very important economic as well as scientifically interesting observations made above would be entirely lost sight of. Furthermore, it would magnify any errors of technic from two to six times or more.

These objections cannot be raised, in like degree at least, to the method of expressing results by reducing percentages to absolute weight per date of each constituent at the several periods of development. It is evident, nevertheless, that this method also can give only a more or less distorted view, because it is practically impossible to select samples with any degree of assurance that they were of the same composition and weight as the previous sample at the time it was

taken. It seems more rational to consider the composition of each sample as we find it, together with its apparent condition of ripeness, the average weight of the individual fruits, the average weight and percentage of seeds, and finally the general relation which the individual fruits forming the sample seem to bear to the whole. By reduction to any other basis, some of these points, especially those involving personal equation in the greatest degree, are masked but not equalized.

Study of the analyses reveals a marked similarity in the composition of the two varieties up to the time of maturity in size. The Deglet Noor shows an inclination toward cane sugar even in the early stages, but from this point on it gains chiefly in cane sugar until ripeness is approached, when some inversion takes place. The amount of this inversion seems to be influenced largely by the temperature to which the ripening date is subjected. Under normal conditions, 20 to 25 per cent. of invert sugar is formed; but under the conditions necessary for artificial ripening, 45 to 50° C for several days, a much larger proportion is inverted. The sample ripened on the tree after frost shows less inversion. The invert sugar date shows an increase in invert sugar parallel to the increase in dry matter. At the period of maximum ripening, when dry matter is accumulating very rapidly, cane sugar appears in considerable quantities. This I attribute to the formation of cane sugar at a greater rate than the invertase is capable of inverting it. The same is also undoubtedly true with other invert sugar fruits, their sugar passing through cane sugar which is at times formed more rapidly than inverted. Many varieties of dates have been examined in this respect, and cane sugar is always observed to accumulate just before the date softens. At the time of softening the invertase, which before this cannot be dissolved by water or glycerin, is now readily extracted by these solvents. In the thick syrupy juice of the date this increased mobility of the invertase must greatly accelerate its action. This accounts for the more rapid disappearance of cane sugar after ripening.

The difference in behavior of the two classes of dates during ripening suggested that the presence or absence of invertase must be the determining factor, and accordingly, the inverting power of a Deglet Noor glycerin extract was tested as follows: 100<sup>cc</sup> of date extract was

added to 2000<sup>cc</sup> of 5 per cent. sugar solution, which was divided into portions of 200<sup>cc</sup> each; one series, containing boiled and unboiled samples, was placed in an incubator at 49° C; the other was left at room temperature. After twenty-four hours no change could be noted in either series. It appeared that Deglet Noor contained no invertase.

A series of comparative extracts of Deglet Noor and three invert sugar dates was then prepared, using five parts of glycerin and seven parts of date, and pressing after digesting together for twenty-four hours or longer. In the tests the same quantities of 5 per cent. sugar solution and of date extract were used in each case. The preservative used throughout was thymol. The figures show the reading on the saccharimeter when portions of the solution were weighed off and treated in the usual manner. It will be noted that the inversion in the case of the three invert sugar dates follows, in a general way, the mass law as found by O'SULLIVAN and THOMPSON<sup>35</sup> for yeast invertase. In another series all the invert sugar date extracts were found to work better at 35° C, but the optimum was not determined.

#### INVERTING POWER OF DATE EXTRACTS

Date	Hour	Deglet Noor	Rhars	Birket el Haggi	Row 12, no. 7
Nov. 6	2 P. M.	5.14	4.51	4.47	4.49
" 6	5 P. M.	5.05	4.32	4.33	4.26
" 7	9 A. M.	5.17	3.36	2.92	3.05
" 7	4 P. M.	5.19	3.01	2.61	2.72
" 8	9 A. M.	5.18	0.81	1.13	1.04
" 9	9 A. M.	5.16	-1.69	-0.65	-1.41
" 10	9 A. M.	5.15	-1.77	-1.99	-1.58
" 12	9 A. M.	5.15	-1.92	-1.79	-1.76

The question then arose as to whether the Deglet Noor contained no invertase or whether an inhibiting substance was present. The first experiment had already shown that the failure to invert was not due to low temperature. Accordingly a series of tests with equal amounts of invert sugar date extract were arranged and increasing amounts of Deglet Noor extract added until 100<sup>cc</sup> of the latter were present to 25<sup>cc</sup> of the former. In no case could a diminution of the inverting power of the invert sugar date extract be observed. To make this matter more certain, a concentrated aqueous extract of

<sup>35</sup> Invertase, a contribution to the history of an enzyme or unorganized ferment. Jour. Lond. Chem. Soc. 57:834. 1890.

Deglet Noor was prepared and found to contain, per liter, 90.32<sup>gm</sup> cane sugar, 18.81<sup>gm</sup> fructose, and 17.78<sup>gm</sup> glucose. A similar artificial mixture was made and both treated with equal amounts of Rhars extract. The natural Deglet Noor extract was inverted slightly faster than the artificial one. This proved that no antifermen was present.

While glycerin extracts of Deglet Noor and M'Kentichi Degla fail to invert solutions of cane sugar, the presence of considerable quantities of invert sugar in these dates points with certainty to invertase. It is untenable, moreover, to consider that these dates differ from the invert varieties merely in quantity of invertase without other qualifications, for a very small amount of invertase will invert a very great amount of cane sugar. O'SULLIVAN found that one part of crude invertase inverted 100,000 parts of cane sugar and still retained its inverting powers. It must be remembered also that a significant amount of invert sugar is formed in a very short time about the period of softening, and that this amount is greater under changed conditions, especially the higher temperature and moisture required for artificial ripening. The inverting action, however, is limited, for cane sugar determinations in samples one year old showed for Deglet Noor 42.14 per cent., M'Kentichi Degla 59.05 per cent., Saffraia 2.96 per cent. The same sample of Saffraia one year before contained nearly 4 per cent. cane sugar. This failure to invert is not due to the invertase becoming inactive, for a Rhars extract made from dates stored one year was fully as active as fresh date extract. A portion of the pulverized Saffraia used above, tested for inverting powers, proved to be one of the strongest preparations examined. These phenomena seem to be due to the localization of the invertase, which means suppression of invertase-secreting tissue. The only thing that could have prevented the inversion of the residual cane sugar in the Saffraia must have been inability to come into molecular contact with the enzyme. The tissues of all dates exhibit inverting powers, although the cane sugar varieties show it but feebly. The failure of their glycerin extracts to invert is probably due to the very small amount of invertase remaining in the same condition as it exists in the green invert sugar date.

Two ways suggest themselves for studying the distribution of the

invertase: testing various portions of the date for inverting power, and comparative analyses of the various parts. Suitable materials for carrying on these lines of work were exhausted this year before a fair start could be made. It is our expectation to continue this phase of the work. Preliminary tests by the first method—dividing the date transversely into blossom third, middle third, and stem third; and concentrically into three parts, the outer shell down to the tannin layer, the tannin layer, and the inner portion—failed to give trustworthy results.

The physiological function of the tannin is little understood. Whatever else it may do, it efficiently protects the fruit from animal ravages and permits it to mature its seed, as has been demonstrated in the Tempe orchard this year.<sup>36</sup> The loss of astringency on ripening was formerly believed to mark the conversion of tannin into sugar, but it was found that in the persimmon<sup>37</sup> the tannin only becomes insoluble. This has been found by THORNBER<sup>38</sup> and the writer to be true of the date also. SLADE considered the tannin present as glucoside which was in some way split up, the tannin being oxydized by an oxydase; and in fact some experiments in which the loss of astringency and softening appeared to have been hastened by the use of manganese salts gave some ground for this opinion. This was further supported by the observations of TICHOMIROV.<sup>39</sup> Even if true, the tannin or tannin-glucoside is so small in amount that it could not appreciably interfere in the carbohydrate relation. Many other quantitatively minor changes undoubtedly take place at the time of softening, among others the change in the behavior of the invertase toward solvents, which I hope to discuss at length in some future paper.

There remains at least one other way in which cane sugar dates differ chemically from invert sugar dates. E. E. FREE, who assisted in these experiments, noticed and called my attention to the fact that sugar solutions to which cane sugar date extracts had been added developed a pink tint. This was more pronounced and developed.

<sup>36</sup> Annual Report, Ariz. Agric. Exp. Sta. 17:164.

<sup>37</sup> BIGELOW, GORE, and HOWARD, *loc. cit.*

<sup>38</sup> *Loc. cit.*

<sup>39</sup> Compt. Rend. 39:305. 1904; through notes of H. B. SLADE.

quicker in boiled solutions than in unboiled, and it was therefore not an enzyme reaction. We found oxygen essential to the color development, because solutions kept in an atmosphere of hydrogen over alkaline pyrogallol never turned pink, even in the bright sunlight, where they were heated to 50 or 55° C. Either in the dark or in the sunlight the color developed readily if free access of oxygen was allowed. This change was never noticed when using invert sugar date extracts. Whether or not it has any connection with the invertase is unknown.

Aside from the chemical classification into cane and invert sugar dates, there is also a physical one into dry and soft dates. We find both cane and invert sugar dates in the dry class as well as in the soft. The Deglet Noor is a typical cane sugar date, yet decidedly a soft date. The M'Kentichi Degla is also a cane sugar date, but of the hard class. The Saffraia, on the other hand, is as typical a hard date as the M'Kentichi Degla, but for all that practically an invert sugar date. The Halloua is a hard but only partial cane sugar date, somewhat similar in sugar content to the artificially ripened Deglet Noor, i. e., about 20 to 25 per cent. of cane sugar. In ripening, the hard dates do not soften or change color; they remain yellow. They shrivel somewhat, but this cannot be used as an index of ripeness, because the shriveling begins before the astringency disappears.

The origin of the cane and invert sugar dates is obscure, and the varieties have existed several hundred years. Nothing is known as to whether they originated from seedlings or were sports. They are now propagated only from suckers. It will prove interesting to study the seedlings to determine whether a cane sugar date can be obtained from a cane sugar date seed. This is an inviting field of investigation, but, due to the slow development of the plant, would require several generations of investigators.